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SCIENTIFIC RESOURCES AND ATOMIC ENERGY

THE fourth annual report of the United Kingdom Atomic Energy Authority*, covering the year ended March 31, 1958, records substantial progress in the Authority's commitments in regard to defence, research and development on the civil applications of nuclear energy, consultancy service to the electricity authorities and collaboration with industry in the nuclear power programme. As was the case with previous reports, it is still impossible to assess the magnitude of the Authority's demand upon the resources of Great Britain in either man-power or materials. The operating account continues to be withheld from publication in the interest of national security, and the balance sheet merely records an item of £2,398,971 for research and development expenditure in aid of design (as against £2,691,341 the previous year). However reasonable that particular course may be, an assessment of the nation's research and development effort as a whole is becoming increasingly necessary, and it would seem from the report that defence considerations are a serious obstacle.

Nevertheless, the report provides a convenient and authoritative review of the Authority's many-sided contribution to Britain's economic development, the value of which is not seriously diminished by the fact that security considerations have been allowed to impose secrecy a little beyond what was essential and thereby hindered discussion of those features of the year's work which have excited most interest and aroused most concern. The time factor may account for the absence of the information which would enable some judgment to be made as to how far the Authority has been able to implement the recommendations of the Fleck reports and eliminate the fundamental weakness which the Windscale accident laid bare, namely, the spreading of the strictly limited amount of skilled staff ever more thinly over a rapidly expanding organization. As regards the future, industrial application of nuclear energy secrecy has not prevented the publication of an outline of the position. Nevertheless, it is important not merely that secrecy should be kept to the minimum but also that there should be general confidence that this is being done.

On this last point the present report adds nothing to the statement of policy given in the previous annual report, and it should be obvious that in the aggregate the functions which the Authority has assumed in regard to education and in its relations with industry must have a considerable effect in disseminating information about nuclear energy, quite apart from publication policy proper and the considerable programme of exhibitions described in this report. The publication policy, including the numerous papers and addresses contributed by the Authority's members and officers, which numbered more than 1,400 during the year, has made much of the information on research and development described in this report familiar to readers of *Nature*, so that recapitulation is unnecessary. It may suffice to note that the uranium situation has eased and that the Authority's interest in helium as a heat-transfer medium has led to drilling at a site in Tanganyika where unusually high concentrations have been reported. The Windscale accident, however, has demonstrated convincingly the value and importance of effective public relations. Indeed, it is perhaps only with that as background that the fullest benefits to all concerned arise from the contacts with industry described in the report under "Relations with Industry".

The Authority can only accommodate a limited number of attached staff, but the hundred or so scientists and engineers from British industry attached to the Authority's establishments at the end of the year are promoting a valuable interchange of ideas between the Authority and industry, acquiring experience for themselves which is not obtainable elsewhere and reinforcing the Authority's own technical resources; no less important, they supplement the contribution to technological education which is made by the Harwell Reactor School and the Calder Operations School, and the five conferences held at Harwell during the year. The Reactor School extended its scope considerably, and the first six weeks of the standard course, which has been increased from fourteen to sixteen weeks, are now held at the Birmingham College of Technology, Bradford Institute of Technology, or Salford Royal Technical College. These courses and the Senior Technical Executive Courses and other courses during the year were attended by 378 British and 120 overseas students, while 32 British and 41 overseas students attended the Calder Operations School, which as more information on the operation of the Calder Hall reactors becomes available is increasingly able to provide a basic course of instruction for those who will ultimately operate similar types of reactor.

The Authority's staff again increased, from 27,290 on April 1, 1957, to 30,341 on March 31, 1958, the industrial staff increasing from 14,660 to 15,860, and reference may be appropriately made here to the Authority's arrangements for training its own staff. These continue to be developed in the light both of the White Paper on Technical Education and of the report of the Carr Committee. In particular, the Authority, which recruited 18 student apprentices and 150 craft apprentices during the year, is reviewing its arrangements in the light of the Carr report, in order to take advantage of the larger number leaving school during the next few years. Training schemes within the Authority, including full-time courses in

* United Kingdom Atomic Energy Authority. Fourth Annual Report for the period 1st April 1957–31st March 1958. Pp. vi+68. (London: H.M. Stationery Office, 1958.) 4s. 6d. net.

laboratory techniques for junior staff, have been further expanded, and full-time degree courses in universities and sandwich courses at technical colleges continue to be awarded both to selected student apprentices and to selected members of the junior staff. Courses and management conferences for senior staff remain a developing feature of the Authority's training arrangements.

This contribution to technological education needs to be kept in mind in considering the demands of the Authority under Britain's man-power resources, while the Authority's internal training programme has a decisive influence in removing what is probably the most serious limitation upon its plans to-day. It is apparent from this report, as from those of the Fleck Committees, how heavy is the responsibility that rests on the works manager and his team at the plant. It is no less apparent that the Authority is seriously short of first-class talent for promotion. Following the reports of the Fleck Committees, the Industrial Group has embarked on an intensive recruitment campaign to bring existing complements up to strength and to fill new posts resulting from the Committees' recommendations. Some of these posts have already been filled by promoting suitably qualified members of the Authority's staff, but the report admits a substantial margin of vacancies to be filled from external sources.

Now while the report quite rightly points out that it is still too early to assess the results of this recruitment campaign, it is fair to comment that there is unlikely ever to be an excess of staff of the high calibre and breadth of experience required for the most senior posts, and the Authority must look in the main for more managers from within its own ranks, and in fact from its own scientists and technologists, who alone are likely to possess the necessary technical qualifications or experience. Moreover, the shortage is not limited to the Industrial Group. There are probably more posts in the higher administration than there are trained men to fill them. The list of senior staff given in this report indicates that the place of deputy director of Aldermaston has not been filled since the former deputy director, Sir William Cook, took Sir Christopher Hinton's place as member for engineering and production.

It is clear enough from this report that the Authority is doing its utmost to increase its staff and to find the men required for the additional posts recommended by the Fleck Committees, though the setting up of the Committee with Sir Douglas Veale as chairman to advise on the training and provision of specialist health and safety staff for the Authority and the nation as a whole, represent in the first instance an increased demand on trained man-power. It will be remembered, however, that the Committee which considered the organization of certain parts of the Authority contemplated deliberate curtailment, if only temporarily, of some of the Authority's activities to enable the Authority to recruit sufficient staff of the high quality required, or to strengthen the technical staff at the levels indicated in the report. It is not apparent from the present report how far the Authority has considered such a reduction in its activities, in order that its programme would match the staff it has available.

Admittedly, there are possibilities that the responsibilities of the Authority may be covered by developments already in train. The new centre of nuclear studies should relieve the Authority of some of the purely physical research, and the universities should be able to do the same as the new equipment now being constructed comes into use. It is reasonable to expect that the relations with industry now being developed could do the same in the development of nuclear power for peaceful purposes; but if the possibilities are to materialize, a deliberate policy of shedding responsibility on the part of the Authority may be required. Some hint that this is being considered might well have been expected in the report, though it is noted that some research in controlled thermonuclear problems is also proceeding in the universities and in the laboratory of Associated Electrical Industries at Aldermaston.

So far as the actual development of nuclear energy is concerned, the report shows that the commissioning of the fast breeder reactor at Dounreay is regarded as less urgent than two or three years ago. The diversion of operating staff necessitated by the Windscale accident has extended the commissioning period, and the fast reactor will not become critical for several months after April 1, 1958, the original date. At Dounreay the Materials Testing Reactor is regarded as a much more urgent requirement, and Sir William Cook has stated that it will be three years before the place of the fast reactor in the hierarchy of industrial reactors can be assessed. Use of plutonium as fuel for fast reactors, in which it is most advantageous, will not be attempted for some time.

Meanwhile, it appears that the Authority's main hopes rest on a further development of the Calder Hall type of reactor, and its primary objective during the 1960's will be to realize the maximum potentialities of the gas-cooled, graphite-moderated reactor system. No further work is to be undertaken on the sodium-cooled graphite-moderated reactor system for the present. Technological and scientific knowledge, it is believed, are growing too rapidly to allow formulation of a firm long-term programme; but the present intention is to use beryllium-clad uranium oxide as fuel, although the reactor will be designed so as to allow alternative forms of high-temperature fuel to be tested on a large scale. The lower capital cost of this advanced gas-cooled reactor will offset the falling load factor which inevitably occurs as nuclear power supplies an increasing proportion of the nation's electricity; and it is possible that a scaled-down version of this reactor system might be suitable for merchant ship propulsion. The Authority is continuing to study the development of alternative systems, however, and by the 1970's work on the fast breeder system might have reached a stage at which commercial power stations can be built. The high-temperature gas-cooled reactor is also receiving detailed consideration as a potential reactor for

nuclear power stations; but the prospects of commercial exploitation of energy from controlled thermonuclear reactions are for the future, and it is as yet impossible to suggest when a contribution to power supplies from this source can be expected.

This recent announcement by the Minister of Supply about plutonium adds interest and importance to the accounts of the processing plants in the Atomic Energy Authority's report, and the decision may well require greater effort at Capenhurst and elsewhere. Moreover, the prospects opened up by the successful navigation beneath the North Pole of *Nautilus* since the report was written might indeed lead to higher priority being given to the work on marine propulsion and possibly justify greater effort in that field. The Authority is already making an intensive study of the engineering design of a merchant ship, which should be complete in about a year; but even if this shows that a merchant ship powered by a nuclear reactor is likely to be economic, such a vessel could scarcely be commissioned before 1963 or 1964.

Studies so far made indicate that a scaled-down version of the Calder Hall type of reactor would be unsuitable for marine propulsion, except for very large vessels, and at present the advanced gas-cooled graphite-moderated reactor is being considered for this purpose as well as a gas-cooled reactor moderated with heavy water, while the organic-liquid moderated reactor system also offers possibilities of a compact system in which the containing vessel need not be capable of withstanding high pressure. Apart from any impetus which may be derived from the achievement of *Nautilus*—and for submarine propulsion the pressurized-water reactor system is particularly suitable—the reactors for marine propulsion, particularly the heavy-water moderated reactor, appear to have potentialities for small land-based power stations and hence for export purposes.

Considerable development effort will be necessary before economic reactors for these purposes can be produced, and the report points out that the diversion of scarce resources, technological skill and development effort cannot be justified until, for example, the prospect is seen of getting to sea a ship that is economic—or well on the road to that end. Nevertheless, the picture may have changed substantially since the report was written and planning permission for construction of the prototypes already granted. What matters, however, is the relevance of that effort not simply to the Authority's own staff and resources but also its relation to the nation's research effort as a whole. The main lesson which the Windscale accident has taught is that the Atomic Energy Authority must adjust its programme to the man-power which it can actually command; hence it is no less essential that the man-power which Britain can put at the disposal of the Authority should be determined with reference to other national needs.

The demonstration of the practicability of submarine polar navigation emphasizes the importance and urgency of one phase of the Authority's work; at the same time, it raises the question whether, when the fullest possible use is made of Anglo-American co-operation and the information which the United States can put at the disposal of Britain, it is not desirable that Britain should put some effort into research and development of the system of inertial navigation. In addition, however, while the Minister of Supply has recently told the House of Commons that the Ministry has been in constant touch with the Royal Society about the possibility of launching an Earth satellite, following his visit to the Woomera rocket range on August 7, Mr. Aubrey Jones has referred to the possibility of Britain undertaking a satellite programme using the Bluestreak long-range ballistic missile and the Black Knight high-altitude research rocket. Mr. Jones admitted that it would be unwise to make such an expensive diversion of effort unless it was warranted by the information that satellites would yield, and he asked the sensible question whether Britain really needs missiles of inter-continental range.

The further question should be asked whether the diversion of effort into the development of an Earth satellite could be justified. The Minister's remarks suggest that serious consideration is being given to a project which has been advocated, largely on grounds of prestige, for almost a year; but Britain can ill afford the diversion of scientific and technological resources, in equipment and materials as well as of man-power and finance, unless there are convincing reasons which would outweigh, for example, any consequent curtailment of nuclear development or research. To launch into such an undertaking except for the most convincing reasons of public interest could be wasteful extravagance.

Less concern might well be entertained over the prospect if there were not so little evidence that, both within the Authority itself and outside it, Britain's effort in the field of nuclear energy is being considered and planned in relation to the total resources of the country and the considered needs of other fields of scientific and technological inquiry and development. There is, on the contrary, much evidence of continuing piecemeal approach and action, and of this the Atomic Energy Authority's report provides further evidence in its silence about the siting of the future nuclear power stations. That is, of course, now the responsibility of the Central Electricity Generating Board; but the Authority might reasonably have been expected to comment on the working life of the nuclear power station. The realization that this is now expected to be no more than twenty or thirty years has deeply disturbed public opinion over the decision of the Minister of Power and the Minister for Welsh Affairs to approve the siting of a station at Trawsfynydd in the Snowdonia National Park and makes the haphazard and piecemeal siting of such stations intolerable.

Suitable sites for large nuclear power stations in Britain are not easy to find; but if such a power station is to be sited in a national park in a country so developed as Britain, there should be irrefutable evidence that the national interest will be grievously and irreparably damaged if the proposal is refused.

Moreover, the obstacles to placing the power station and its transmission lines underground should be made public; and if cost is the sole obstacle, that should be precisely known.

It is the failure to face the implications of a considered policy and to recognize that Britain's resources in finance, man-power, materials, equipment and amenity are limited and can be stretched too far, which is responsible for the concern over Government policy in regard to technological development. The Atomic Energy Authority's report supplies no evidence that the period of piecemeal action is ended. Accordingly it is necessary to reiterate that implementation by the Authority itself of the recommendations of the Fleck Committees alone will not suffice. If Britain is to reap the full benefits of the varied activities of the Authority—benefits which extend not merely into the medical field but increasingly, with the opening of the Wantage Radiation Laboratory, into such fields as industrial polymerization and the improvement of synthetic fibres and reduction of the immense loss of stored products through insect pests—it is not enough to bring the Authority's own programme and activities into balance with its resources in trained man-power. It is no less essential that the resources made available to the Authority are themselves in harmony with the other numerous and conflicting demands on the nation's limited resources of finance, materials, equipment and trained man-power.

MECHANICAL TRANSLATION

Mechanical Resolution of Linguistic Problems

By Dr. Andrew D. Booth, L. Brandwood, and Dr. J. P. Cleave. Pp. vii+306. (London: Butterworths Scientific Publications; New York: Academic Press, Inc., 1958.) 50*s*.; 9.50 dollars.

Programming for an Automatic Digital Calculator

By Dr. Kathleen H. V. Booth. Pp. vii+238. (London: Butterworths Scientific Publications; New York: Adademic Press, Inc., 1958.) 42*s*.; 9.80 dollars.

THE second of these books is a companion volume to the former, and is essentially a programmers' handbook to the APEXC, the magnetic drum computer at Birkbeck College, London, which has been used for many of the experiments on language translation described by Dr. Booth and his colleagues. The book is not only a companion volume, of course, but also contains much information about how the more familiar processes of numerical analysis are programmed for a machine of this type. Thus, for example, the topics covered include input and output, division and square root, evaluation of functions by power series, floating point operations, solution of simultaneous linear equations, evaluation of latent roots, and finally, 'post mortem' checking programmes.

The first book deals primarily with language translation, although by way of introduction the authors include as "linguistic problems" various aspects of the investigation of literary style, such as lexicography, and the analysis of rhythm and syntax. For all these purposes the text has to be encoded and punched on teleprint tape (or other input medium) for scanning by the machine. The first four chapters describe how the machine can read and interpret information presented in this form. The question of stylistic analysis is discussed in relation to the problem of determining the chronological sequence in which Plato wrote his works.

In Chapters 5 and 6, there is a very good account of the general aspects of language translation, and the programming techniques used to overcome some of the difficulties. Thus it would appear that the first task is to construct a stem-ending dictionary so as to reduce the vocabulary to a manageable size, otherwise if all the inflected forms of every word were given the dictionary would grow to unmanageable proportions. The dictionary look-up process matches each word of the input text with a dictionary of 'stems'. When the stem has been located it is removed from the word and the remainder looked up in a dictionary of "endings". The two pieces of information derived from these dictionaries together give the equivalent of the input word. Naturally, variations of this procedure are necessary for a language in which inflexions are made by means of prefixes. Refinements of the matching procedure are also necessary in the case where the stem and ending are connected by redundant joining letters (as an example the authors consider some regular French verbs). To operate a dictionary in this case necessitates left-to-right matching to select from the stem dictionary the longest entry which is part of the input word. Once the stem has been removed, right-to-left matching is used to locate the ending. The remaining letters can then be discarded. A similar technique is used to deal with compound words, which are such a common feature of technical literature.

The dictionary search procedure which the authors recommend for an automatic computer is essentially the same as one would normally use, that is to say one opens the book (roughly) in the middle, and then proceeds to narrow down the entry in question by a partitioning process. By employing a binary subdivision process for this purpose a dictionary of N entries can be searched in approximately $\log_2 N$ matchings.

Other problems considered in Chapter 6 are connected with word order, idioms, multiple meaning and ambiguity. The problem of word order, for example, is to translate "équation différentielle est" into "differential equation is". A fairly simple procedure suffices in this case, to invert the order of adjective and noun, but in translating from highly inflected languages, where the relations between words is governed almost entirely by endings, more elaborate devices are necessary to ensure the appropriate rearrangement in the English equivalent.

Chapter 7 is concerned with the mechanical transcription of Braille, a problem which at first sight appears to have little to do with language translation. The difficulty, however, lies in the rules for forming *contractions*, where parts of words, such as 'con' and 'ion', are abbreviated to a single Braille symbol. While clear enough in their intention, they are not immediately suitable for programming because they cannot always be expressed formally in terms of the sentence structure, but depend very much on the context of the symbols the contraction of which is to be made; for example, they should not overlap well-

defined syllable divisions. The authors conclude that the machine production of Braille text is not an economic possibility at present, in view of the large amount of voluntary effort available.

Chapter 8 describes an experimental scheme for translating from French into English devised for APEXC. For reasons of storage space the scheme was limited to a dictionary of 250 words and their English equivalents, but the programme was sufficiently comprehensive to perform certain operations of syntactical nature, and to recognize the masculine and feminine, singular and plural forms of nouns and adjectives, and also all parts of the verb except the first person imperative and the subjunctive mood.

The last part of the book is devoted to the translation of German, a subject which, unfortunately, the reviewer is not able to comment on.

The authors conclude by considering the technical details of a machine designed specially for translating. The principal feature of such a machine, of course, is a large high-speed store. Various special orders are recommended to facilitate the dictionary look-up process, and at the same time orders such as multiplication can be dropped. The authors suggest that such a machine could be built for between £50,000 and £100,000 but are rather sceptical of obtaining financial support on this scale. However, their proposed instruction code for a linguistic machine is not so very special, and it would scarcely seem worth while to build a machine on this scale which would exclude it from doing tasks other than language translation. The authors' best hope, therefore, lies in the super-high-speed computers at present under development, such as Stretch or Larc.

R. A. Brooker

WORK ON FUSED SALTS

Soviet Research in Fused Salts (1949–1955)

(Chemistry Collection, No. 1.) General. Pp. 28. Structure and Properties: Ternary Reciprocal. Pp. 200. Structure and Properties: Quaternary Systems; Quaternary Reciprocal Systems. Pp. 98. Structure and Properties: Preparation; Iodine Systems. Pp. 156. Structure and Properties: Binary Systems: Ternary Systems. Pp. 284. Slags and Mattes. Pp. 39. Electrochemistry. Pp. 54. Thermodynamics. Pp. 58. Translated from the Russian. (New York: Consultants Bureau, Inc.) n.p.

INTEREST in the structure and properties of fused salts has been growing with noteworthy vigour in the past few years in various countries. From the fundamental point of view, molten salts constitute a distinct class of fluids, with an extensive range of temperatures over which the liquid state may be stable, with high heats of vaporization, and with quasi-crystalline structures in which the tendency for an ion of one sign to be preferentially surrounded by ions of opposite sign imposes an additional element of order, compared with other liquids. Despite the technical difficulties to be overcome, these characteristic properties have stimulated various basic researches on the transport and thermodynamic properties of ionic melts, and on melting mechanisms of ionic crystals.

The very great importance of fused salts in electrometallurgy, particularly of the more electropositive or reactive cation-forming elements, has for many years prompted applied research the bulk of which greatly exceeds the body of fundamental studies available in published literature. Much of this research is somewhat empirical. In addition to their interest for electro-metallurgy, fused salts have many analogies with fused silicates used as slags in bulk production of various metals. Their physical properties for applications as heat transfer fluids and bath fluids are also attracting increasing attention. All these considerations make any additions to published research, especially on the fundamental properties of fused salts, particularly welcome at the present time. Unfortunately, for many Western scientists researches published in Russian must still be regarded as not freely available until translations become accessible. The appearance of this collection prepared by the Consultants Bureau Inc. can therefore be described as very timely.

Translations necessarily suffer from certain defects in reproducing new knowledge. For example, the present collection includes nearly one hundred papers on melting and phase relationships for binary, ternary and quaternary mixtures of salts. The curves by means of which much of this information is recorded suffer in precision through the process of reproduction; such records may not be sufficiently accurate or clear for some purposes. However, it is very useful to know even the approximate shape of the diagrams in many cases.

In addition to a great bulk of work on phase diagrams, a limited number of the papers included give calculations on activity coefficients from electromotive force data, on the surface tension, density, viscosity and electrical conductivities of melts, on corrosion of metals by melts, and on slags.

Though discussions of basic theoretical considerations are included in some of the papers, the inspiration of much of this work appears to be technological. It constitutes an extensive body of factual knowledge about fused salts which should prove useful to those who do not have access to the original publications.

A. R. Ubbelohde

SOCIAL MEDICINE

The Teaching of Hygiene and Public Health in Europe

A Review of Trends in Undergraduate and Post-graduate Education in Nineteen Countries. By Prof. F. Grundy and Prof. J. M. Mackintosh. (World Health Organization: Monograph Series, No. 34.) Pp. 254. (Geneva: World Health Organization; London: H.M. Stationery Office, 1957.) 15 Swiss francs; 25*s.*; 5 dollars.

DURING the past hundred years scientific methods have been applied widely in medical research and practice; in the same period the health of the population of the Western world has improved. But the improved health owes less to advances in medical science than to changes in the external environment, and a favourable trend in the standard of living. We are healthier than our ancestors not because of what happens when we are ill but because we do not become ill; and we do not become ill not because of specific protective therapy but because we live in a healthier environment. In its preoccupation with the minutiae of diagnosis and the pathogenesis

of disease, medicine is in danger of neglecting what has hitherto proved its most powerful resource—the manipulation of the external environment—and of failing, when it can do no more, "au moins à nous mener à la fin de la vie par un chemin agréable".

These are among the considerations which have led to an attempt to improve the teaching of preventive and social medicine, and the report prepared for the World Health Organization by Prof. F. Grundy and Prof. J. M. Mackintosh provides a valuable account of the instruction given in the medical schools of Europe. They describe briefly the curriculum in different countries, and interpret the trends in undergraduate and postgraduate teaching. Perhaps the most obvious conclusion suggested by their report is that there are still wide differences of opinion about what is needed. In Oslo and Amsterdam it is considered desirable to have two separate departments of hygiene and social medicine, and this pattern is soon to be extended to Sweden. In London no medical school has a chair of social medicine, and instruction is entrusted to part-time teachers or to other departments.

There are equally wide differences of opinion about the interpretation of the subject itself, for while the concept of social medicine evidently means a great deal to many people, it does not always mean the same thing. To the original core of traditional public health teaching have been added a number of other subjects—medical administration, medical statistics, human genetics, demography, social biology etc.—some of which have little more in common than the facts that they are obviously important, and either are not taught or are inadequately taught by any other department. Indeed social medicine has come to be identified with almost any medical theme which is statistical in its method or social in its implications; it is scarcely surprising that the syllabus which has resulted is overweighted and lacks coherence. As Grundy and Mackintosh recognize, if the subject is to make its full impact on medical education there will have to be some contraction of its ill-defined terms of reference, with, one would add, a synthesis of the material which remains.

Thomas McKeown

POLES, NORTH AND SOUTH

Arctic and Antarctic

A Prospect of the Polar Regions. By Colin Bertram. New edition, entirely revised. Pp. ix+123+13 plates. (Cambridge: W. Heffer and Sons, Ltd., 1958.) 21*s*. net.

NINETEEN years ago, in the preface to the first edition to this book, the author wrote, "The idea then came to me that it might be worth while attempting, not to record the narrative of an expedition, but to state some principles, physical, physiological and psychological, which govern the life of men in polar lands, presenting them in a way which might be of interest to others than specialists. When the attempt was made to follow out this plan, it soon became obvious that the underlying principles of dietary, housing and transport could be far more attractively presented in a setting that depicted the life itself". The result was a new type of introduction to the polar regions. It was based on wide reading and personal experience in both the Arctic and the Antarctic, it gave an enormous amount of information in a few pages, and it was well written. Now, with the added qualification of seven years as director of the Scott Polar Research Institute, Dr. Bertram has brought out a second, and greatly revised, edition of his book.

The history of polar exploration he divides into three stages: the pioneer stage before the First World War, the intermediate stage between the World Wars, and the present modern stage. The progress from stage to stage is "largely to be measured in increasing complexity and enlarged material resources. The simple motive of personal high endeavour . . . has been transformed into national political aspirations".

He begins with a general survey of the polar regions and the animals, birds and fish on which travellers there may exist. Few species occur in the Arctic, and still fewer in the Antarctic, but such as there are congregate in numbers seldom seen in the rest of the man-infested world. Insects and penguins are as numerous as ever, but other species have been all but exterminated at one time or another by man for economic gain, and only survive now through protective legislation. Then the main aspects of polar life are taken in turn and their progress through the three stages, pioneer, transitional and modern, is examined; they are physiology, acclimatization and clothing, dogs, transport, food, organization and leadership. These chapters contain so much that is interesting that extracts are impossible; certainly one would need to be a voracious reader to collect all these facts elsewhere.

Dr. Bertram's long and varied experience of polar affairs makes his appreciation of the "Polar prospect to-day" particularly valuable and interesting to anyone concerned with these regions. The International Geophysical Year of 1957–58 he sees as a turning point in Antarctic history. In spite of the scientific motives that have taken so many nations south there is little doubt but that political changes will result. New sovereign areas, agreed spheres of influence or internationalization, each is possible; all that the author feels certain is that "political factors will largely prevail over the claims of international law as at present understood".

It is a pity that this book has appeared so late in the recent stream of polar books, and may meet with undeserved neglect on this account.

L. M. Forbes

HOW NEUTRONS GET THERE

Neutron Transport Theory

By B. Davison, with the collaboration of J. B. Sykes. (International Series of Monographs on Physics.) Pp. xx+450. (Oxford: Clarendon Press; London: Oxford University Press, 1957.) 75*s*. net.

IN the construction and the performance of nuclear reactors, problems connected with the movements and distribution of neutrons inside the reactors are of prime importance. Some of these problems were solved even before neutrons were discovered, in the setting of astrophysics. The current theory of neutron transport pays due homage to this venerable parentage by much of its terminology and techniques. On the other hand, young as the subject is, it has already outgrown its ancestry by the subtlety and variety of the problems and the demand for the great accuracy in the solutions. Most of the (published) progress of the subject dates from the later years of the Second

World War and onwards; the author of the book under review has a distinguished and well-known record in that progress. He has now produced in a massive and carefully written volume a fine account of the various mathematical methods which have so far been brought to bear on the problems of neutron transport. In his task he received the active co-operation of a number of eminent specialists in the field. Readers who approach this subject with experience in some other branch of physics will be interested, possibly surprised, to read in the book the contributions to transport theory by physicists (for example, Feynman, Fuchs, Pryce) more widely known in other fields.

The book starts with a general statement of the problems involved: this may be equivalently framed either as the Boltzmann equation or as an integral equation. In the greater part of the rest of the book various solutions, analytical and numerical, are considered. The reader is left in no doubt as to which method to apply in a particular situation. The ordering of the treatment is, quite appropriately, to take simpler problems first and consider others in an order of increasing complexity. The result is that roughly the first one-third of the book is 'compulsory reading', after which various sections in the remainder are independent of each other. The intention of the author to make the mathematical treatment detailed and self-contained has been achieved with remarkable feats of economy.

The book is primarily directed to the advanced worker in neutron transport theory. To him the book offers mainly the following three advantages: the opening chapters contain a balanced and precise justification for the physical assumptions of the theory. Secondly, by presenting the subject as a systematic and compact discipline this book promises to be a standard one, as regards form and notation, and as regards the inter-relation of topics treated in the book. Lastly, much of the material is published in this book for the first time.

This book can be strongly recommended to those interested in other branches of transport theory. It is not always that the methods developed in connexion with neutrons can be legitimately applied; nevertheless, there is a great deal to be gained from familiarizing oneself with these methods.

It is a pity that the book makes scarcely any reference to the way in which these elaborate calculations have been tested or put to use. Now it may seem misguided to ask for this kind of information, much of which is probably not at the author's disposal, in what is primarily a text-book of methodology. Nevertheless, one hopes that future circumstances will enable the author to make allowance for a (proverbially deadly) weakness of his reader.

R. Englman

MINERAL REQUIREMENTS OF PLANTS AND ANIMALS

Mineral Nutrition and the Balance of Life

By Dr. Frank A. Gilbert. Pp. xv+350+30 plates. (Norman, Oklahoma: University of Oklahoma Press, 1957.) 5.95 dollars.

ANIMAL and plant physiology have an important link in the mineral nutrition of the plant. Many instances are now known, and others are constantly coming to light, where a deficiency in a food plant has important consequences on animal health. These discoveries have led to greater emphasis on the problems of interrelationships between plant and animal nutrition, and this in turn has intensified investigations on the mineral content of soils of different localities. It is perhaps not surprising that nutritional disorders of animals due to deficiency or excess of minerals were not immediately associated with the mineral composition of the food plant because elements essential to animals are not necessarily essential to plants, and the latter have the facility of accumulating elements which take no part in metabolism but have no harmful effects on their growth. Thus, cobalt is not considered essential for higher plants but the small amount that occurs in plants is of great importance because normally this is the only way in which animals get their essential supplies of this element. Some plants are tolerant to the presence of selenium in the soil, but such plants are toxic to animals.

The story of mineral deficiencies in plants is in itself a fascinating one and Dr. Gilbert has not only given a very complete account of the mineral requirements of plants but also linked it with the question of nutrition and well-being of animals that use these plants as food. The treatment of this subject may be said to be comprehensive since the author considers each element in turn, first from a historical point of view and then how the lack (or in some cases excess) of that element affects plants and animals, including humans. These accounts are linked to the relation between deficiencies and the soils of particular localities with mention of the geographical regions where deficiencies prevail. It is inevitable that most of the latter information refers to the North American continent, where the most intensive work has been done, but available information from other continents is included.

So far about sixty elements have been identified in plants, but only about twenty of these are conceded to be essential either to plant or animal nutrition. The present book, however, deals with thirty-one, including not only the major elements nitrogen, phosphorus, potassium, calcium and magnesium and the trace elements but also others such as aluminium, silicon, selenium, fluorine, arsenic and lead which are of interest by virtue of their toxic properties.

The author has included a chapter on enzymes, hormones and vitamins which seems out of place in a book of this nature. The brief chapter on analytical methods underlines the fact that the book is not primarily addressed to the specialist although there is much in the book which is of interest to those concerned with mineral nutrition and the 1,177 references are a valuable guide to the literature, but by no means exhaustive. The book concludes with a chapter on human nutrition which repeats the main theme of the book that the health of the animal depends on a supply of nutritious forage. The author points out that we are protected by law against harmful drugs but not against inferior food, and pleads for more education and publicity to create a realization of the need for better quality food.

The book is well produced and contains numerous photographs illustrating symptoms of mineral deficiency and toxicity in plants and animals. It can be recommended as a good general account of the mineral requirements of plants and animals.

E. C. Humphries

Selected Combustion Problems, 2

Transport Phenomena; Ignition; Altitude Behaviour and Scaling of Aeroengines. M. W. Thring (Senior Editor). (AGARD Combustion Colloquium, Liège, Belgium, December 5 to 9, 1955.) Pp. viii + 495. (London: Butterworths Scientific Publications; New York: Interscience Publishers, Inc., 1956.) 90*s*.; 12.50 dollars.

THIS is the record of the combustion colloquium, held at Liège in December 1955. Eighteen papers were presented and discussed at some length, the languages used being English and French. Abstracts are rendered in both tongues with occasionally interesting results, and a small subject index of doubtful value is appended.

The papers are, as the main title of the book suggests, on chosen aspects of combustion. The choice naturally has regard to the interests of aeronautical scientists, and the sub-title rather incompletely indicates group headings. The author's aim is generally to survey, and little new experimental work is presented.

There is no doubt that colloquia of this nature are of value in assessing the current state of knowledge and in defining those problems that most clearly call for study. It is good to see evidence that time was allowed for extended discussion.

As to how far the record is of permanent value on the library shelf is another matter. A possible view is that the practical success of the meeting will be measured by the speed with which the papers and discussion become out-dated. Meanwhile, those who are concerned with aero engines (including rocket motors) and their fuels, fundamentally considered, will find ample material for consideration within these pages. J. H. Burgoyne

The Alimentary Tract of the Ruminant

By David Benzie and Dr. A. T. Phillipson. Pp. 24 + 54 plates. (Edinburgh and London: Oliver and Boyd, Ltd., 1957.) 27*s*. 6*d*. net.

INVESTIGATION of the gut of the ruminant during the past twenty years has made the chemical changes which go on within the stomach reasonably plain. This is not, however, true of the mechanical activity of the alimentary tract and there is still a considerable degree of doubt as to the way in which food is sorted into that fraction which goes to the rumen and that fraction which is passed along the œsophageal groove to the abomasum, and as to the muscular movements which play a part in the belching which these animals use in order to get rid of the gas produced during ruminal digestion. A relatively new approach to these problems has been made with the use of radiographic studies, a series of which is reported in this volume. Part of the work was done in the Department of Physiology of the Rowett Research Institute, and part in the Department of Anatomy at the University of Manchester. The animals appear to withstand the effect of radiography well and not to be much affected by this experimental method. The book consists largely of an atlas of 54 plates, most of which contain several X-ray photographs of the alimentary tract of young animals (lambs, calves, kids) fed a barium meal. The movements of the gut are obviously complex, but among the interesting points which the series of photographs makes plain is the confirmation of the extra-ruminal contraction described by Weiss as the basis of the belching mechanism. Since the work was carried out predominantly on young animals mainly fed on milk, the part played by the chambers of the stomach other than the abomasum is not made plain, though an interpretation of the activity of the omasum is given. There is every expectation of this most promising method leading to clarification of the muscular movements carried out by each part of the stomach in the alimentary tract of these animals. A. Graham

Structural Psychology

De Humani Mentis Fabrica. By D. and K. Stanley-Jones. Pp. viii + 180. (Bristol: John Wright and Sons, Ltd., 1957.) 21*s*.

THE authors of this book define structural psychology as "a system of psychology erected upon the basic sciences of anatomy and physiology" (page v) and further state that "the contribution which structural psychology has to make to the major problem of philosophy has been to prove that the structure of the mind and the structure of the brain are sufficiently alike in every known point of comparison to warrant the statement that they are identical. By proof of the identity of structure the hypothesis is justified that there is an identity both of location and substance" (p. 160).

It is suggested that emotion originated in palæolithic times when man lived mainly in caves, and these were conditioned to "lust and feeding in the warmth of the night, aggression in the outside cold of the day". Aggression is associated with oral biting and massive discharge of the orthosympathetic system, and in balanced antagonism with oral sucking and the parasympathetic system. Obstruction of the sucking leads to a mass discharge of the orthosympathetic with oral biting.

These basic reactions become associated with the mother, envisaged by the child as the bad mother when it is unsatisfied and the good mother when it is. From her the emotions are transferred to the father and so to society.

It is impossible to describe the whole of the authors' arguments in a few words. They link physiology ingeniously with psychological concepts, but their system is not completely new and is mainly a fresh arrangement of Freudian psychology and organic theories. It should prove interesting to those psychologists interested in physiology and those physiologists psychologically orientated.

The book is clearly printed on non-shiny paper, the style is lucid and should be understood by educated readers even if unfamiliar with the material. There is an excellent index and glossary.

Intermediate Physics

By Dr. C. J. Smith. Fourth edition. Pp. xiii + 1322. (London: Edward Arnold (Publishers), Ltd., 1957.) 45*s*. net.

OVER some twenty-five years the successive editions of this standard intermediate text-book have proved their value. There are 300 more pages than in the 1947 edition, and the extensive rewriting to clarify points which students always find difficult, with the addition of material to bring it well up to the requirements of a good scholarship candidate, will certainly increase even further the number of people who will use it happily and with success. Noteworthy additions are the fuller treatment of

dynamics and of the experimental side of electrostatics, and a chapter on forced vibrations and resonance in the section on acoustics. There are also many more numerical examples for solution.

Dr. Smith holds strong views about the MKS system, which one gathers he would not touch with a pole; and his comment on units in the preface may seem a little sweeping. His mastery of the orthodox approach in the sections on electricity, and the general excellence of its presentation, nevertheless compel one's admiration. G. R. NOAKES

Principles of Engineering Heat Transfer

By Prof. Warren H. Giedt. Pp. xiii+372. (Princeton, N.J., and London: D. Van Nostrand Company, Inc., 1957.) 48*s*. net.

IN view of the extensive literature of the subject, the number of text-books on heat-transfer is surprisingly few and makes this new addition especially welcome. The author has rightly devoted the major part of the work to a fundamental exposition of the basic principles, avoiding embroilment in the jungle of empirical data which has grown up with the development of this subject. The book follows the natural division of heat-transfer into the three modes of conduction, convection and radiation, all of which are treated with admirable clarity. The order of treatment is, however, somewhat unusual, the separation to opposite ends of the book of the chapters on steady-state and transient conduction appearing to serve no useful purpose. The relegation to the last chapter of the concept of logarithmic mean temperature difference, which is so fundamental in the study of heat-exchange equipment, is also a less desirable feature of the book. Convection is preceded by two chapters on fluid flow which in the main are most helpful to an understanding of convective heat-transfer principles. There are, however, certain superfluities for a book of this kind, the most noticeable of which is the inclusion of the elementary theory of orifice and venturi meters.

Among the commendable features of the book are the summary of the principles of the heat and momentum transfer analogy in the solution of turbulent convection problems, including the extension to the special problem of liquid metals, and the description of electric analogue methods in the solution of conduction problems.

The book will be extremely useful to the student at university degree-level but would have been improved by the inclusion of numerical answers to the many end-of-chapter problems, a deficiency which, alas, is a common failing of almost every text-book of American origin. H. COHEN

Die Bluteiweisskörper des Menschen

Untersuchungsmethoden und deren klinischpraktische Bedeutung. Von Prof. Ferdinand Wuhrmann und Dr. Charlie Wunderly. Dritte völlig umgearbeitete Auflage. Pp. 499. (Basel und Stuttgart: Benno Schwabe and Co., 1957.) 52 Swiss francs.

THE previous two editions of this treatise, published in 1947 and 1951, were, perhaps, much better known on the Continent than in Britain because no English versions were available. This edition is an extensive revision and enlargement of the former editions. A welcome feature is the prominence now given to paper electrophoretic and agar gel immunoelectrophoretic techniques. Here, at last, is a book which brings together so many illustrations of abnormal plasma protein patterns. Another feature is the thorough discussion of the traditional techniques of free electrophoresis, ultracentrifugation and salting out. Routine clinical methods for blood examination are also well covered; the clinical significance of the plasma proteins, their evaluation and their alteration in disease are comprehensively discussed.

There is a tendency, perhaps understandable, to refer more to German than to non-German sources, but consequently some important work is overlooked or not given the attention it deserves. Recent techniques of zone electrophoresis in columns or slabs or of ion exchange chromatography are not mentioned though they have been used with striking success for plasma protein separation. In such a detailed book the subject index might perhaps be fuller. However, there is a good balance between the physico-chemical and clinical sides and in this respect the book is exceptional. Each of the authors is a specialist in each field and they have collaborated for many years in clinical investigations. Though rather expensive the price is not too high for such an attractively printed, comprehensive reference work. Its appeal would undoubtedly be widened if an English version was available. R. CONSDEN

Selected Lectures in Modern Physics for School Science Teachers

Edited by Prof. H. Messel. Pp. 168. (Sydney: Nuclear Research Foundation within the University of Sydney, 1958.) 30*s*.

IN January of this year the Nuclear Physics Foundation of the University of Sydney held a summer school for science teachers. This was not just a refresher course to attack or remove the outer layers of rust. Its expressed intention was to infect the participants with an inspiration which they would spread, and the direct aim a second-order effect on the future entrants to the faculties of science and engineering. This record of the proceedings, which was made available at the end of the school (in itself a remarkable feat of publishing), is both a worthy tribute to the venture itself and an excellent survey of many fields of modern physics.

The twenty-four lectures range widely, and the topics seem to have been chosen for their general interest; they are the matters on which a teacher would very properly be expected to be able to answer questions. Nuclear energy; low-, high-, and ultra-high energy nuclear physics; 'strange' particles; physics in rain-making; Newton's laws of motion and space-travel; radioastronomy; how to talk to an electronic brain; transistors; the use of isotopes in medicine; physics in geology; measurement; and cosmic radiation; these are among the main subjects selected. There are also an instructive account of the building of relatively simple apparatus for demonstration experiments in electronics and nuclear physics, and a discussion of careers in physics, and of the need of suitable candidates for them.

The lecturers themselves are to be congratulated on touching just the right level for their audience, and on communicating their obvious enjoyment of their work. I learned a great deal from the book, felt that I understood most of it, and was much impressed by the way it was all presented. The book can be highly recommended to teachers of science, and will be a most valuable acquisition to a school science library. G. R. NOAKES

THE LYOT $H\alpha$ HELIOGRAPH AT THE CAPE OF GOOD HOPE

By Dr. M. A. ELLISON

C.S.A.G.I. Reporter for Solar Activity, Royal Observatory, Edinburgh

THE organization of the International Geophysical Year has provided a splendid impetus to the study of solar – terrestrial relations. Quite early in the planning stages it was realized that the solar chromosphere must be kept under continuous observation throughout the eighteen months. This meant the provision of equipment, well distributed in longitude and located in the most favourable climates, which would automatically photograph the Sun in hydrogen light at 1-min. intervals. In addition, the observing periods at existing stations, equipped with visual spectrohelioscopes, had to be planned in advance on a time basis so as to fill in the gaps and avoid duplication[1].

This organization has become known as the 'solar patrol'. Some 25 stations are now filming the Sun, and by their combined efforts it is hoped that no solar activity which may be of significance in the study of the ionosphere, geomagnetism, cosmic rays or solar radio emission, will pass unobserved.

In the middle of 1955, the British National Committee for the International Geophysical Year decided to make a substantial contribution to the work of the solar patrol. A sum of £17,000 was voted for the provision of a Lyot-type heliograph along with some auxiliary apparatus and the building to house it. The site chosen was the Royal Observatory, Cape of Good Hope, where under Her Majesty's Astronomer (Prof. R. H. Stoy), expert staff and maintenance facilities would be available. The sunshine hours at the Cape average 68 per cent of possible, compared with some 40 per cent in the most favourable part of the British Isles. For many years the Cape Observatory has contributed daily photographs of the Sun in white light to the Greenwich photoheliograph programme; and in 1949 the Sun was photographed at the Cape on 360 days.

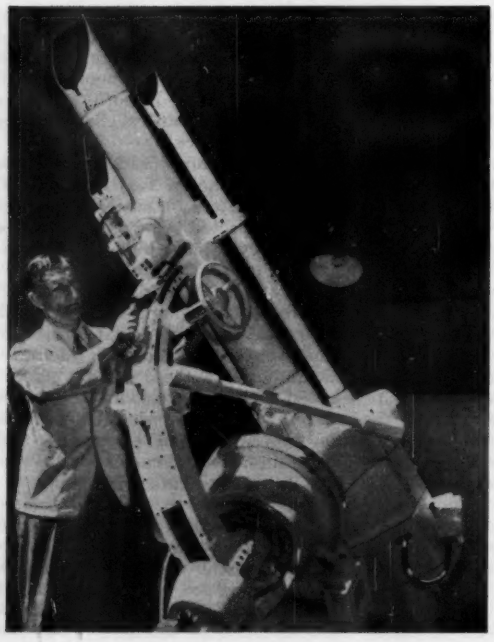

Fig. 1. Lyot $H\alpha$ heliograph installed at the Royal Observatory Cape of Good Hope

The contract for the construction of the heliograph was placed by the Admiralty with the firm Société d'Études et de Construction d'Appareillages Scientifiques et Industriels of Bordeaux in December 1955. The completed instrument was accepted there in November 1957 and delivered to the Cape on February 25, 1958. After successful tests of the optics and electronic circuits, it was brought into daily operation on March 16.

We owe the original conception of the monochromatic heliograph to the late Bernard Lyot. After his lamented death in 1952, the plan was brought to a practical outcome by the construction of a prototype instrument at Meudon, chiefly through the labours of the d'Azambujas and Dr. A. Dollfus. The production on a commercial basis was undertaken by the Société d'Études et de Construction d'Appareillages Scientifiques et Industriels, the study group of which was responsible for the equatorial mounting, the electronic and electro-mechanical arrangements, as well as the auxiliary and cinematograph optics. The main objective and the monochromatic filter were constructed by the firm Optique et Précision de Levallois.

The instrument comprises three separate optical trains. In the centre (Fig. 1) is the monochromatic telescope, consisting of an objective of 14 cm. aperture working at f 10. The beam from this lens traverses two glass prefilters before being rendered parallel for its passage through the Lyot filter. This filter is of the standard design[2], being built up of eight birefringent plates of quartz and others of calcite with intermediate 'Polaroids', all contained inside a cylindrical block of aluminium. The temperature of the block is thermostatically controlled so that at the working temperature of the filter (44·2° C.) the band pass of 0·7 A. is maintained precisely fixed upon the centre of the $H\alpha$ line (6562·8 A.). On emerging from the filter, the monochromatic beam is focused on the film frame of the camera placed in the breech of the telescope and forms there a solar image 15 mm. in diameter. Exposures are made on standard 35-mm. film; using Kodak Panchromatic Microfile emulsion, the normal exposure time is 2 sec. The duration between exposures may be pre-selected for intervals of 60, 30 or 15 sec. By the operation of a uni-selector switch every eleventh frame receives an exposure time five times normal. This has the effect of over-exposing the disk so as to record only the prominences at the Sun's limb. An eye-piece conveniently placed at the side of the camera allows the image in $H\alpha$ to be observed visually in the intervals between exposures.

The second optical train (right-hand tube of Fig. 1) is that of the electronic guider and exposure meter. The main drive to the equatorial in right ascension is provided by a synchronous motor which operates

through the usual reduction gear, worm and driving wheel. The electronic guider compensates for all errors in the main drive, for maladjustments of the polar axis and for refraction changes at varying altitudes. The image to be photographed is thus maintained centred in the film frame with an accuracy of 1–2 sec. of arc —an important feature when the films are to be examined later with a cinematograph projector.

The lens system of the guider forms a solar image about 1 in. in diameter. This image illuminates a wide slit tangential to the east limb together with a circular hole placed some distance inside the west limb; the slit has an aperture twice the area of the hole. When the light flux through each is the same, the photocells behind receive equal illumination and their currents cancel one another. An excess of current in either direction, due to a lateral shift of the image, operates a symmetrical valve amplifier the output of which affects a magnetic amplifier with two stages. This, in turn, operates in either sense the diphase motor of the right ascension slow-motion. A similar arrangement of apertures and photocells, placed at right angles to the former pair, corrects for errors in declination. Such a system has the added advantage that it functions independently of the seasonal variations in the apparent diameter of the solar image.

Fig. 2. Heliograph exposure, April 7, 1958, 10h. 55m. 54s., showing a Class 3 flare in operation. Normal sky orientation

In the same optical system the centre of the solar image illuminates a small aperture behind which is placed a fifth photocell. This cell acts as a photometer and regulates the duration of each exposure. The start of the exposure is obtained by applying a negative charge to the condenser of a light-integrator, which, by means of a valve *6AK5*, releases the trigger of the camera shutter. The condenser is progressively discharged by the cell of the photometer. When the condenser has discharged, the same valve again becomes conducting, pulling the shutter trigger and terminating the exposure. The exposure-times are thus proportional to sky opacity, which means that the resulting solar images on the film are of nearly uniform density, a most important feature for accurate photometry.

When thick cloud intervenes, a 'no light' relay comes into operation, switching off the camera action in order to conserve film. As the sky clears, the image is automatically re-centred and the operating cycle begins again.

The third optical system (to the left of the main tube in Fig. 1) serves two purposes. First, it contains a clock and calendar which are illuminated by diffuse sunlight. The clock is set to Universal Time at the start of each day's run, and its image, focused on the film by means of a lens-cum-prism train, records the time and date of each exposure (Fig. 2). Secondly, an echelon of eight circular spots is focused below the clock in order to provide a photometric calibration. The echelon is formed of eight blocks of neutral glass, each having been ground and polished to a suitable thickness so as to provide density steps of 0·1. These blocks are uniformly illuminated by direct sunlight which has previously traversed a red filter and an opal diffusing screen. Thus the quality of the light forming the calibration image on the film is similar to that of the solar image, while the exposure-time is the same for both whatever the transparency of the sky.

The equatorial mounting is of unusual design, having been planned with a short polar axis and a wide fork to combat the effects of flexure and wind vibration. Rising from the lower end of the polar axis are two telescopic arcs of radius 80 cm. which are concentric with the declination axis. The extension of these arcs by means of a rack and pinion provides for the seasonal variations of $\pm 23°$ in declination. The inner arc carries at its head the diphase motor and screw of the declination slow-motion, the upper end of the telescope tube being held in contact with the screw by means of two extensible springs. The telescope thus receives support from the three angles of an isosceles triangle.

A control cabinet for the instrument houses under damp-proof conditions the filter thermostat, the time-relays for operating the exposures and all the electronic and electro-mechanical parts. Controls are effected by press-buttons on the front panel, where indicator lamps also enable the operator to judge the correct functioning of the various circuits. Experience gained at the Cape fully supports the claim for automatic operation. Apart from the routine procedures of setting on the Sun, regulating the clock and loading the film cassettes, it performs its daily run without further attention until close-down. The roof of the building has been so constructed that a clear aperture extending across the sky is available at all seasons.

The films taken with the heliograph (about 25 ft. per day) are being developed at the Cape. They are then dispatched to the Royal Observatory, Edinburgh, where they are projected and examined for the occurrence of flares and blow-off prominences, the records of these phenomena being contributed monthly to the International Geophysical Year World Data Centres. The images on 'Microfile' film can be enlarged to a diameter of 6 in. before the film grain becomes noticeable; and on this scale they

show considerably greater resolution of fine detail than the best spectroheliograms.

The high performance of the filter is also illustrated by the low intensity of the scattered light. On a day of moderate sky transparency, the scattered light (sky plus instrumental) at a distance of 2 min. of arc outside the solar limb was measured as 3 per cent of the intensity of the central disk. With most spectrographs the scattered light would have five times this value. On the other hand, the intensity at the centre of the disk in the $H\alpha$ image was found to be 28 per cent of the intensity of the continuum outside the wings of $H\alpha$. This is high compared with the known value of 15 per cent for the centre of the $H\alpha$ line; it arises from the relatively wide band-pass of the filter (0·7 A.), which causes it to transmit appreciable light from the line wings.

One of the most important investigations to be undertaken with the Lyot heliograph will be the photometry of solar flares. Accurate light-curves for these events are required so that we may be in a position to compare them with the development of: (1) the sudden disturbances in the terrestrial ionosphere, for example, geomagnetic crochets, radio fade-outs and sudden enhancements of atmospherics, which occur almost simultaneously; and (2) the 'outbursts' of radio emission which are generated in the flare regions. In Fig. 3 are plotted photometric light-curves for three distinct regions in a Class 3 flare which was recorded with the heliograph on April 7 (see also Fig. 2). One can observe here how the regions *A* and *C*, which were separated by 98,000 km. in the chromosphere, brightened up and reached their maximum intensity simultaneously. The development of the sudden ionospheric disturbance (recorded as a sudden enhancement of atmospherics on frequency 24 kc./s.) is shown in the same diagram. The time-lag of 12 min. between the peak intensity of the flare and that of the sudden ionospheric disturbance confirms the similar results previously obtained from visual records of flare line-widths made with the spectrohelioscope[3].

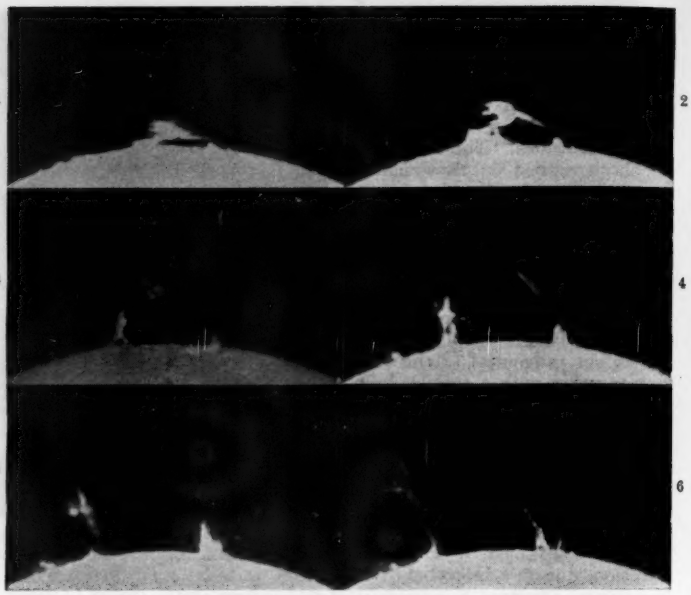

Fig. 4. Blow-off prominence on the west limb, March 14, 1958. Times: (1) 14h. 39m. 10s.; (2) 14h. 50m. 15s.; (3) 14h. 54m. 15s.; (4) 14h. 58m. 25s.; (5) 15h. 01m. 25s.; (6) 15h. 04m. 25s.

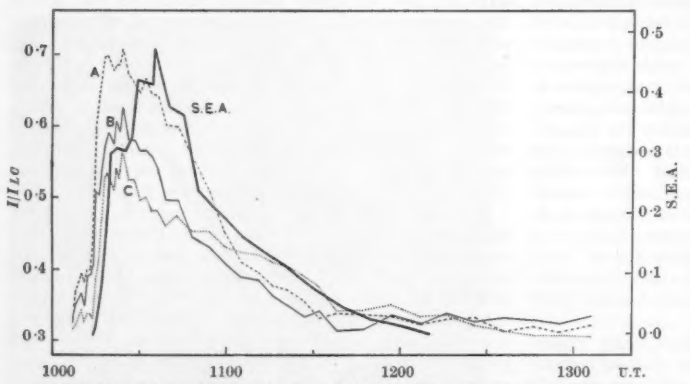

Fig. 3. Photometric light-curves plotted for three regions in the Class 3 flare of Fig. 2. Intensities are expressed in terms of the local continuum outside the wings of the $H\alpha$ line. The heavy line shows the development curve of the sudden ionospheric disturbance (recorded at Edinburgh as a sudden enhancement of atmospherics on a frequency of 24 kc./s.)

Lastly, in Fig. 4 we can see the great potentialities of the instrument as a means for providing a rapid sequence of pictures of prominence material blowing off into space at the Sun's limb. The top of this prominence arch rose through 180,000 km. in 19 min.; before it faded away, it was moving with a velocity of 625 km./sec. across the line of sight. Had the blow-off occurred near the centre of the disk, instead of at the limb, the sightline component of velocity would have shifted the prominence $H\alpha$ line some 14 A. to the violet, right outside the pass-band of the filter. The mechanism which leads to this sudden outward acceleration of solar material remains unsolved.

Three similar Lyot heliographs have been completed and delivered by the Société d'Études et de Construction d'Appareillages Scientifiques et Industriels. One has gone to Tokyo, another to Washington and the third to the McMath-Hulbert Observatory, Michigan. In the coming years we may therefore anticipate much fuller records leading to a more complete understanding of chromospheric activity; a happy situation that has been brought about largely through the inspiration and the funds of the International Geophysical Year.

[1] Öhman, Y., *Ann. Int. Geophys. Year*, **5**, Part 4 (1958).
[2] Dollfus, A., *Rev. d'Optique*, **35**, 539 (1956).
[3] Ellison, M. A., *J. Atm. and Terr. Phys.*, **4**, 226 (1953).

LONG-RANGE TRAVEL OF THE RADIOACTIVE CLOUD FROM THE ACCIDENT AT WINDSCALE

By N. G. STEWART and R. N. CROOKS
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LOCAL air measurements made during the course of the accident on October 10, 1957, at Windscale No. 1 Atomic Pile showed that radioactivity was released into the atmosphere between noon on October 10 and noon on October 11 (ref. 1). Meteorological conditions throughout this period were complex. During October 10 a ridge of high pressure extended over England from the east, giving mainly light south to south-west winds. A shallow depression passed close to Windscale during the evening of that day, causing the local winds to back to an easterly direction and become very light for a few hours, after which the wind returned to its original south-westerly direction. During the day, the air in the lowest 400 ft. of the atmosphere was unstable, with an inversion above. By late evening, the air near the ground was stable.

Early on October 11, at about 0100*Z*–0200*Z*, a cold front, which during the previous day had been moving south-eastwards across Scotland, passed over Windscale. At the passage of the front the wind quickly veered to the northerly and increased to about 10 knots and slowly backed to north-west during the next twelve hours. The front reached London about 1330*Z* and cleared Britain by about 1900*Z* on October 11. Behind the front, the air was unstable up to about 4,000 ft., but by the evening of October 11 the lowest layers of the atmosphere had again become stable.

Cloud Sampling

As the radioactive cloud passed over the United Kingdom and Europe, samples of the airborne dust were collected on filter papers which had been exposed in many places for monitoring industrial smoke or the dust from nuclear test explosions. Most of these filters had been exposed for 24-hr. periods, but at a number of sites continuously moving strip filters had been used, and these provided a detailed picture of the passage of the cloud overhead.

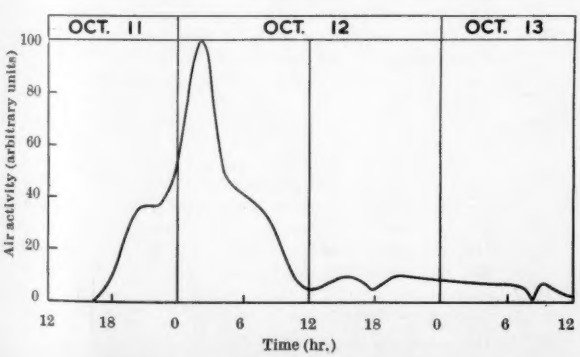

Fig. 1. Time variation of air activity at Harrow, Middlesex

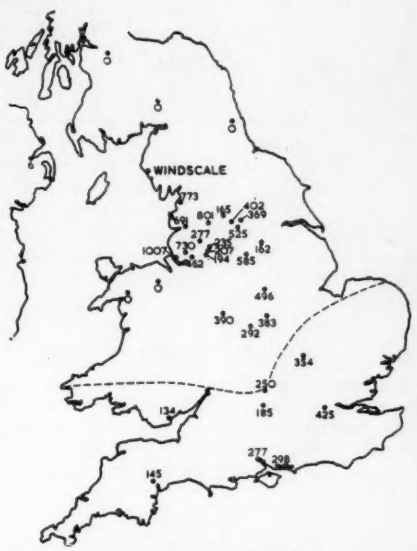

Fig. 2. Dosage of iodine-131 in air ($\mu\mu c$-days/m.^{3})

The majority of the samples from Great Britain were obtained from the public health departments of local authorities and from the Fuel Research Establishment by Mr. G. B. Courtier, of the Scientific Branch of the London County Council, who sent them to Harwell for detailed analysis. Many of the samples from Europe were obtained through the chairman of the International Geophysical Year Advisory Committee on Nuclear Radiation, Prof. W. Bleeker, who arranged for the filters exposed at routine sampling stations to be forwarded to Harwell for the measurements to be made on a common apparatus.

The results show that the cloud travelled ultimately in a south-easterly direction and had substantially passed over Lancaster by noon on October 11. It is possible to deduce from the 24-hr. samples in the Leeds–Bradford area that the cloud first reached there at about 0900*Z* on October 11 at the same time as the cold front, suggesting that this part of the cloud had initially travelled in a north-easterly direction on October 10 and had been afterwards swept southwards with the wind accompanying the front. The arrival of the cloud at Harrow, Middlesex, at 1600*Z* on October 11—some three hours after the cold front—is demonstrated in Fig. 1, which is taken from a record obtained by Dr. Herz, of Kodak, Ltd., using a moving-strip filter apparatus. The main part of the cloud passed over in about 20 hr., but low levels of activity persisted for

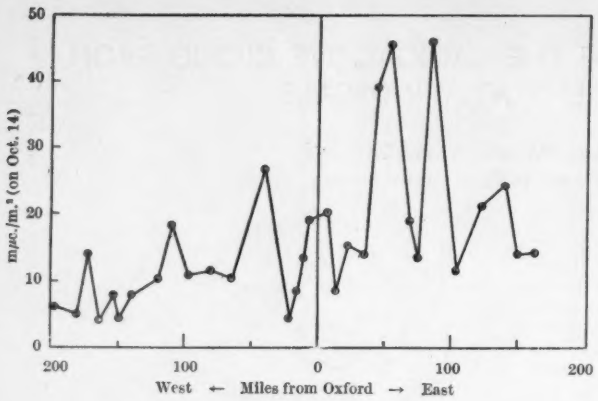

Fig. 3. Survey of iodine-131 on grass

another three days. The cloud first reached Mol (Belgium) at 1900Z on October 11, Frankfurt at 2200Z on October 12 and Sola (Norway) at some time on October 15.

Analysis of Radioactivity

The nature of the radioactivity was analysed radiochemically and by gamma-ray spectrometry, and the relative amounts of the main components on a filter exposed at Harwell (360 km. from Windscale) throughout the time of passage of the cloud are given in Table 1.

Table 1

Isotope	Half-life	Relative activity at 0001 hr. on October 11
Iodine-131	8 d.	100
Tellurium-132	77 hr.	85
Iodine-132	2·3 hr.	85
Cæsium-137	30 y.	11
Ruthenium-103	40 d.	7
Polonium-210	135 d.	2·6
Strontium-89	54 d.	0·8
Strontium-90	28 y.	0·04

The figures differ in some respects from those quoted by Chamberlain and Dunster[2] for a short-period air sample at Calder. In particular, the ratio of iodine-131 to cæsium-137 on the Calder filter was 38·5 as compared with 9·1 at Harwell. The mean value at ten sites in Great Britain south of Lancaster was 9·0 ± 1·8 (standard deviation).

The concentration of iodine-131 in the air at each sampling point has been integrated over the time of passage of the cloud to give cloud dosage figures expressed in μμc.-days/m.³. These have been plotted for the United Kingdom in Fig. 2, which shows that

Table 2

Place	Iodine-131 dosage (μμc.-days/m.³)
Paris	2·7
Brussels	49
Eindhoven	31
Hanover	8·8
Potsdam	2·7
Vienna	1·0
Zug (Switzerland)	0·2
Hradec Kralove (Czechoslovakia)	small
Sola (Norway)	1·9

the cloud avoided north Wales but diffused to cover a wide front in the southern part of the country. The degree of dispersion is better shown in Fig. 3, which presents the results of a survey of the iodine-131 deposited on grass between St. David's and Cromer (dotted line in Fig. 2). It is clear that the dust cloud spread out into the North Sea and it was probably this part of it which reached Mol in Belgium some three hours after the first radioactivity reached London. The iodine-131 cloud-dosage figures observed in Europe are, as expected, smaller than those found in southern England, and some representative values are given in Table 2.

The significance of the iodine-131 cloud dosages may be judged from the fact that at the highest recorded dosage-level in Fig. 2, a child would have inhaled about 0·01 μc. of iodine-131; this is about 1 per cent of the amount it would have ingested by drinking milk at the control-level defined by the Atomic Energy Authority and afterwards ratified by the Medical Research Council[1].

Deposition of Radioactive Material

Little rain fell while the cloud passed over England, and the radioactive dust was deposited on grass mainly by turbulent diffusion and impaction. The relation between the concentration of any isotope in air and the amount deposited can be expressed in terms of a 'deposition velocity' defined by:

$$V_g = \frac{\text{amount of isotope deposited per unit area}}{\text{time integral of air concentration } (= \text{dosage})}$$

Values of V_g were determined at places where air and grass measurements were made reasonably close together. The mean value for iodine-131 obtained near Preston, Burnley and Sheffield was V_g = 0·30 cm./sec., while that in southern England was 0·11 cm./sec. These values are lower than the experimental figure of 2·5 cm./sec. obtained experimentally by Chamberlain and Chadwick[3,3] at Harwell under adiabatic conditions but are qualitatively consistent with the stable air conditions which obtained during the passage of the cloud. The velocities of deposition of cæsium-137 and ruthenium-103 were less than 15 per cent of that of iodine-131; a similar difference between iodine-131 and other fission products has previously been observed by Megaw and Chadwick[4]. This preferential removal of iodine-131 from the cloud would be expected to affect the isotopic composition of the residual dust, and it is significant that the ratio of iodine-131 to cæsium-137, corrected for radioactive decay, decreased from its mean value of 9·0 in England to 5·2 in Germany and to 2·8 in Norway, where the cloud did not arrive until October 15.

We wish to acknowledge the co-operation of the many people in various European countries who collaborated in this investigation. We would also like to thank our colleagues at Harwell who analysed the samples, especially Miss E. M. R. Fisher and Mr. D. L. O. Humphreys.

[1] Accident at Windscale No. 1 Pile on 10th October, 1957. Cmnd. 302. (H.M.S.O., 1958.)
[2] See succeeding article.
[3] Chamberlain, A. C., and Chadwick, R. C., *Nucleonics*, **11**, 8 (1953).
[4] Megaw, W. J., and Chadwick, R. C., *A.E.R.E., HP/M* 114 (1956).

DEPOSITION OF RADIOACTIVITY IN NORTH-WEST ENGLAND FROM THE ACCIDENT AT WINDSCALE

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AND
H. J. DUNSTER
U.K. Atomic Energy Authority, Risley

Emission of Activity

THE course of events during the accident to No. 1 pile at Windscale on October 10 and 11, 1957, has been described in the report of the Committee of Investigation under Sir William Penney[1].

On the afternoon and evening of October 10 there was calm weather with light variable wind. A cold front was approaching from the north-west. In advance of the front there was a south-westerly wind which had an estimated speed of 7 knots at 500 ft. This wind blew for about three hours only, and with the passage of the front at Windscale about 0100 hr. on October 11, it was succeeded by a stronger northerly wind backing north-west, which blew for the remainder of that day.

The pattern of fall-out along the Cumberland coast shows that most of the deposition occurred after the north-west wind became established. Since the emission was from a 400-ft. stack, and in the presence of hot gases, the degree of turbulence at different times, as well as the wind direction, was important in determining the areas of deposition of activity.

The materials readily available for determining the nature and pattern of the deposition of activity were filter papers through which air had been passed for monitoring purposes during the incident, and grass which had received fall-out activity. Areas of one or two square yards were closely cut from areas such as roadside verges not subject to grazing by cattle. The method of measurement mostly used was gamma spectrometry. The samples were weighed and mixed, and aliquots of about 100 gm. wet-weight placed in polyethylene bottles on the crystal of the spectrometer under reproducible geometrical conditions. It was thought undesirable to dry the grass before estimating the iodine activity, lest some be lost by volatilization, although in fact no such loss was observed when samples were left exposed to the air. The dried weight of some samples was estimated by drying other aliquots. Calibration for the various nuclides was achieved by adding known amounts of activity to similar materials, and counting them under the same geometry. To enable the amounts of nuclides other than iodine to be measured, the analysis of many samples was repeated during December, by which time the iodine-131 had decayed. Details are given elsewhere[2].

Of the minority of fission products which do not emit gamma-rays, strontium-89 and strontium-90 were of obvious importance, and radiochemical analyses for these isotopes were performed.

In the first column of Table 1 are shown the activities of various nuclides, relative to iodine-131, on an air filter operated from 1000 hr. to 1030 hr. on October 11, at Calder Works, 800 metres south-east of Windscale Pile No. 1. Five cubic metres of air were drawn through the filter, and the iodine-131 activity on it was 5 μc. Also in Table 1 are shown the relative activities corrected to October 11, on samples of grass taken on October 13 at Seascale and Drigg, 3·2 and 6·4 km., respectively, south-east of Windscale. The iodine-131 activity of the Seascale sample extrapolated to October 11 was 0·16 μc./gm. dry-weight, and that of the Drigg sample 0·04 μc./gm. dry-weight. These results are similar to those of Kimber and Booth[3], who found 0·08 μc./gm. dry-weight in grass from a point 4·8 kilometres south of Windscale.

Table 1. ACTIVITY RELATIVE TO IODINE-131 OF FILTER PAPER AND GRASS SAMPLES

Nuclide	Air filter (Calder)	Grass	
		(Seascale)	(Drigg)
Iodine-131	100	100	100
Cæsium-137	2·6	1·2	2·0
Strontium-89	1·0	0·2	0·2
Strontium-90	0·02	0·004	0·01
Barium-140	2·5	—	0·5
Ruthenium-103	3·8	1·5	1·7
Zirconium-95	0·3	0·2	0·3

Small traces of other isotopes such as cerium-141, cerium-144 and ruthenium-106 were also detected.

The nuclides listed in Table 1 do not include those of short half-life; for example, tellurium-132 and its daughter product iodine-132. The presence of short-lived activities contributed appreciably to the gamma dose-rate immediately after the incident, but they were not important from the biological point of view.

Not all the fission products present in the grass were due to the Windscale incident, since all samples contain activity due to fall-out from distant bomb tests. Foliar contamination from this cause is thought to account for a considerable fraction of the activity of the non-volatile fission products such as zirconium-95 found in the grass.

The degree to which the more volatile fission products were enhanced in the activity emitted is illustrated by the ratio of iodine-131 to strontium-89. In long-irradiated fuel these nuclides are in the ratio 100 : 160, so that Table 1 shows that the escape of strontium-89 was less than 1 per cent of that of iodine-131, relative to the amounts in the pile.

Radioautographic examination of filter paper and other surfaces exposed to fall-out showed that most of the radioiodine was in gaseous form or absorbed on very small particles. Negligible amounts of uranium were disseminated in the accident, but some particles believed to be graphite with absorbed fission products were found both on air-filter papers and on the ground close to the pile. An extreme example was a 15-micron particle containing 0·8 μc. of barium-140, 0·4 μc. of strontium-89 and 0·01 μc. of strontium-90, with their daughter products, but no other detectable fission product activity. There

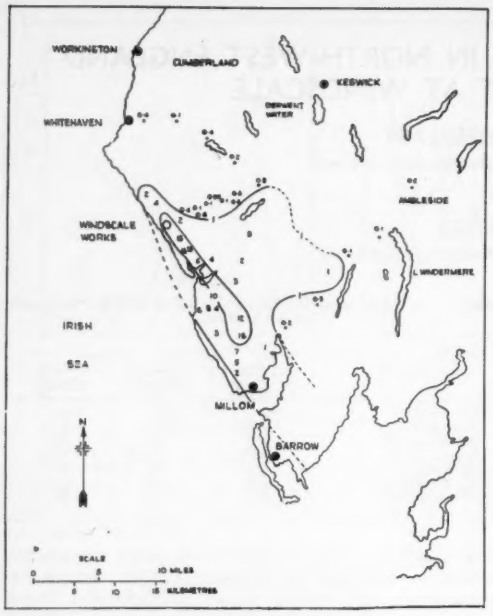

Fig. 1. Contours and spot-levels of iodine-131 activity on grass in $\mu c./m.^2$ on October 11, 1957

evidently had been a physical separation of the elements barium and strontium, the boiling points of which are 1,140° and 1,150° C., respectively, from other fission products.

Other particles were found with activity due largely to cæsium-137. The activity associated with particles distinguishable by autoradiography or by the survey of surfaces with beta counters was, however, small in comparison with the total activity on the surface, and this was still true when measurements were repeated after the iodine-131 activity had decayed.

Deposition of Iodine-131

The fall-out pattern in Cumberland is shown in Fig. 1. The iodine-131 activity of grass has been worked back to October 11 by using the results at a location where repeated samples were taken. These showed that the activity in cut samples expressed in microcuries per square metre ($\mu c./m.^2$) decayed with an effective half-life of 5 days due to the combination of radioactive decay and other factors. Many more results are available than can be inserted in the figure[2].

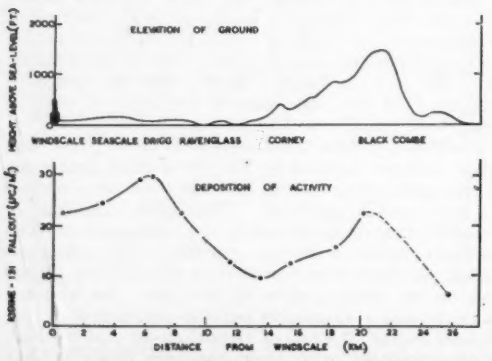

Fig. 2. Deposition of iodine-131 as a function of distance and elevation along line bearing 145° from Windscale

There is a narrow strip along the coast in which the contamination of the grass initially exceeded 10 $\mu c./m.^2$, but the 1 $\mu c./m.^2$ contour extends to the Westmorland border and into the Barrow peninsula.

Theoretical relationships have been established between the amount of iodine-131 deposited and the dose-rate from the gamma-rays which are emitted[5]. Using these relations, measurements of gamma dose-rate have been used to derive estimates of the iodine-131 activity per unit area of ground, with results that agree quite well with the direct measurement of the activity of the grass. For example, at Seascale the iodine-131 activity in grass on October 11, from the sample of Table 1, was 15 $\mu c./m.^2$, whereas the activity deduced from the gamma dose-rate at a nearby point was 17 $\mu c./m.^2$.

The deposition of iodine-131 along the axis of the fall-out plume south-east from Windscale, estimated in this way, is shown in Fig. 2. There is a maximum of fall-out at Drigg, 6 km. from Windscale, and another on the slopes of the mountain Black Combe, 18 km. away. The second maximum was probably due to the elevation of the ground and the associated turbulence in the air over it, which would increase the transfer of activity to ground level.

Biological Implications

The results reported here were obtained with the aim of evaluating the physical processes affecting the released radioactivity. The greater part of the survey work following the accident was, however, concerned with assessing the magnitude and extent of the danger to man. This work was concentrated on human foodstuffs such as milk, eggs, meat, drinking water and vegetables; and it confirmed the conclusions reached from previous experimental work and theoretical assessments that iodine and strontium isotopes in milk would be the limiting contaminants. In fact, the preferential release of iodine relative to strontium made iodine the only material of biological significance.

An empirical correlation of the activities of milk and grass showed that about 10 $\mu c./m.^2$ of iodine-131 on grass was required to give 1 $\mu c.$/litre of milk. In order to protect children, milk supplies were condemned for human consumption in an area where any of the samples showed contamination of 0·1 $\mu c.$/litre or more. The limit of this area followed roughly the 1 $\mu c./m.^2$ contour of Fig. 1. It was not found necessary to impose any restriction on other foodstuffs.

A more detailed report of the surveys of milk and other foodstuffs will be published separately[4].

The collection and analysis of samples was done by many members of the staff of the Atomic Energy Authority both at Windscale and Harwell. We are especially grateful to H. Howells, W. L. Templeton, F. J. Woodman and A. E. Ross at Windscale, and to D. V. Booker, F. J. Bryant, W. H. Hardwick and M. J. Heard at Harwell.

[1] Accident at Windscale No. 1 Pile on 10th October, 1957. Cmnd. 302. (H.M.S.O., 1957.)
[2] Booker, D. V., *A.E.R.E. HP/R* 2607 (1958).
[3] Kimber, G., and Booth, A., *Nature*, **181**, 1392 (1958).
[4] Dunster, H. J., Howells, H., and Templeton, W. L., paper to Geneva Conference, *A/CONF.* 15/*P*/316 (to be published, 1958).
[5] Chamberlain, A. C., *A.E.R.E. HP/R* 2606 (1958).

THE BRITISH ASSOCIATION FOR THE ADVANCEMENT OF SCIENCE

New President

SIR JAMES GRAY, professor of zoology in the University of Cambridge, has been elected president for 1959 of the British Association for the Advancement of Science in succession to Sir Alexander Fleck.

Sir James has had great influence on the course that zoology has taken in Great Britain. All who were concerned with the development of the experimental approach to the study of animals which occurred in the years following the First World War would regard James Gray as the leader, and his books on ciliary action and on experimental cytology as milestones along a broadening road of achievement. The interest in movement revealed in the former led to enrichment of knowledge in a series of admirably planned and beautifully executed researches on locomotion in segmented worms, in fish and in higher vertebrates.

Early in his history he assumed the editorship of *The Journal of Experimental Biology*, which he held for thirty years, establishing it as a major scientific journal and coming in this capacity to guide the work of a whole generation of younger workers. His achievements at Cambridge are there for all to see, not least in the high distinction of his colleagues in the Department. The Marine Biological Laboratory, Plymouth, owes much to him, from the stimulus of his presence as a young research worker to the culmination of influence as president of the Marine Biological Association. He is a trustee of the British Museum, has served on the Agricultural Research Council and on many other bodies, but with little doubt would put first among these his continuing responsibilities as a Development Commissioner and as the chairman of its Fishery Advisory Committee. Few with his distinction in the field of fundamental research have encountered more widely the implications of science on human welfare.

New Officers

Lord Rennell of Rodd succeeds Mr. Maurice G. Bennett as general treasurer of the British Association. Lord Rennell's scientific interests are mainly geographical and in 1922 and 1927 he carried out explorations in the Sahara. He was president of the Royal Geographical Society in the immediate post-war period 1945–48 and did much to revive and expand the activities of the Society. He has been largely responsible for organizing the appeal for additional funds on behalf of the British Association, and the measure of his success is obvious in the number of new fields in which the Association is now operating.

Dr. Norman Wright has been elected biological general secretary of the Association in succession to Dr. George Taylor. Dr. Wright is chief scientific adviser (food) to the Ministry of Agriculture, Fisheries and Food, and he is distinguished for his contributions to research in animal ecology and the application of nutritional science to human welfare. He was the first director of the Hannah Dairy Research Institute, Ayr, where he was charged with determining the design and layout of the new buildings and initiating the research programme. He is a member of a number of important committees concerned with food and agricultural matters and has served on the Agricultural Research Council. He has travelled widely on advisory missions. At the invitation of the Governments, he has reviewed the cattle and dairy resources of India, Pakistan and Ceylon, and also the animal industry in the Middle East. He is chairman of the Programme Committee of the Food and Agriculture Organization in Rome and in 1952 he was president of Section *M* (Agriculture) of the British Association.

Soddy Commemoration Plaque

The Soddy Commemoration Plaque in the Chemistry Department of the University of Glasgow was unveiled on August 27. Sir Hector Hetherington, vice-chancellor and principal of the University, presided at this occasion. He welcomed some relatives of Prof. Soddy who were in attendance and Prof. H. J. Emeléus, who represented the Royal Society. He then read a complimentary telegram from the Nobel Foundation, Stockholm. Sir Alexander Fleck, with a wise, reminiscent and affectionate speech, unveiled the bronze plaque with its portrait in relief and the citation "With Rutherford he shared the discovery of atomic disintegration, with Ramsay the proof of helium production from α-particles. In this Department he formulated his conception of isotopes". Prof. J. Monteath Robertson, in accepting the memorial tablet, thanked Mr. James Barr, the artist, and also Dr. Andrew Kent, who had organized the proceedings and also a Soddy exhibition including the original piece of isotopic lead—208—which had been loaned by Prof. McNeill of Toronto.

FREDERICK SODDY, M.A., LL.D., F.R.S.

A photograph (R. Cowper) of the plaster-cast by James Barr, D.A., for the memorial plaque in the Chemistry Department, University of Glasgow. It is based on photographs of or about his Glasgow period (1904–14) and on the Cannizzaro medal by "T.S.S.".

Endeavour Prizes

The prizes awarded for the 1958 *Endeavour* Essay Competition were presented by Sir Alexander Fleck on August 29, during the reception for exhibitioners in the Randolph Hall of the University of Glasgow. The competition is sponsored annually by Imperial Chemical Industries Limited, publishers of the quarterly scientific review *Endeavour*, and is limited to those under twenty-five years of age. Ninety-two entries were received, but, although many were from abroad, all the prize-winners this year come from the British Isles. The first prize of 50 guineas was awarded to Mr. Charles Perrin, of Winchester College, for an essay on "Darwinism Today". The second prize of 25 guineas was awarded to Miss Angela Glynn for an essay on "Education for an Age of Science". Miss Glynn is a student at Newnham College, Cambridge. The third prize of 15 guineas was awarded to Mr. John Christopher Horrocks, of Halifax, Yorkshire, for an essay on "Rockets and Satellites and their Scientific Significance". Mr. Horrocks is studying for an external London degree at Bradford Institute of Technology. Two special prizes of 5 guineas were awarded to competitors under eighteen. Mr. Anthony Orchard, of Swansea, received one of these for an essay on "Chemicals from Petroleum". He is at present at the Bishop Gore Grammar School for Boys. The second was won by Mr. Andrew Billson, of Nottingham, for an essay on the same subject. Mr. Billson is at the Becket School, West Bridgford.

NEWS and VIEWS

Physics at the University College of Wales, Aberystwyth : Prof. W. J. G. Beynon

Dr. W. J. G. Beynon, at present a senior lecturer in the University College of Swansea, has been appointed to the chair of physics at the University College of Wales, Aberystwyth, in succession to the late Prof. R. M. Davies (see *Nature*, 181, 879 ; 1958). Dr. Beynon graduated in the University of Wales (University College of Swansea) in 1934, with first-class honours in physics, and then proceeded to research in the Department of Physics at Swansea, obtaining his Ph.D. degree for work on magneto-optical dispersion in 1939. During 1938–46 he worked in the Radio Division of the National Physical Laboratory on ionospheric problems in radio communication. In 1946 he returned to the Department of Physics at Swansea as lecturer, being promoted to senior lecturer in 1949. With research students, he continued work on ionospheric problems, and in 1951 was awarded the degree of D.Sc. (Wales). His published work is concerned with radio-wave propagation, reflexion and absorption in the ionosphere, and in some papers he collaborated with Sir Edward Appleton. Recent publications have been on solar eclipses and radio studies during the International Geophysical Year, for which he is a 'world correspondent' on the ionosphere. He has been general editor of the Proceedings of the International Joint Commission on the Ionosphere, and has served on many committees, national and international, including those dealing with artificial Earth satellites, and rocket investigation of the upper atmosphere. He was a member of the official British delegation to General Assemblies of the Scientific Radio Union at Zurich (1950), Sydney (1952), The Hague (1954), and Boulder (1957), and has recently visited Moscow.

Report on Education

The report of the Ministry of Education, "Education in 1957" (pp. v+198. (Cmnd. 454). London : H.M. Stationery Office, 1958. 9*s*. net), brings together a good deal of information which has been given piecemeal to Parliament, and presents a convenient picture of trends, some of which, notably in further education, are of particular interest to the scientist. During 1956 the number of senior children in all-age schools was reduced by 10 per cent, while during 1957 completion of new buildings permitted the re-organization of 203 all-age schools, and there are 260 projects in the approved building programme, completion of which will enable 1,300 more all-age schools to be re-organized. New building work to the value of more than £60 million has now been authorized for the first four years of the five-year plan for the expansion of technical education, out of a total of £70 million, and by the end of 1957 work had begun on twenty new colleges and 102 major extensions to existing colleges. Although the staffing position in the primary schools improved slightly, it continued to deteriorate in the secondary schools, and the report emphasizes the need for a considerably larger number of teachers of mathematics and science when the number of children in the secondary schools rises in the next few years. Statistics and a graph included in the report point to a new peak expected in the school population in 1968 almost as high as that expected in 1961. The number of full-time teachers in technical colleges rose from 10,817 to 12,139, and of part-time teachers from 47,024 to 49,320, but the increase is still short of that considered necessary by the Willis Jackson Committee. The year was notable for the number of visitors to the Science Museum, the figure of 1,317,000 having only been exceeded in 1935 (1,327,000).

Russian Scientific Journals available in English

The U.S. National Science Foundation has published a useful list of Soviet scientific and technical journals now available in English translation. 60,000 pages a year are now in print and these include 53 English editions of Russian journals, four extensive series of translated Russian abstracts of scientific papers, and four series of partial translations of important Russian journals. The list includes subscription rates and details of where the translations can be ordered. It can be obtained from the National Science Foundation, Washington 25, D.C.

The Australian Museum, Sydney

So long ago as 1827 Earl Bathurst suggested that a public museum be established in New South Wales, where "many rare and curious specimens of natural history are to be procured". By 1852 the first portion of the permanent building had been erected. The whole building will be completed by the construction of the North Wing, which is due to begin very soon. From its early days, the Museum has been dis-

tinguished for arranging its exhibits in attractive form and for publications designed to appeal to all with an interest in natural history. The latest publication, "Handbook to the Australian Museum", replaces the former "Guide" and has been designed to help make visits to the Museum of increased interest by providing information not only about the exhibits themselves but also of the theme that links them all together. Approximately 250,000 people visit the Museum each year.

The report of the Trustees of the Australian Museum for the year ended June 30, 1957 (pp. 23. Sydney: The Australian Museum, 1958), includes a valuable summary of the growth and development of this progressive institution. The main purpose of this statement is to indicate that for a period of no less than forty-seven years no major building developments have taken place, and this in spite of many applications to the authorities for increased accommodation. Plans and designs for new displays have been prepared for the aboriginal, bird, fish, invertebrate, insect, mineral, mammal and fossil galleries. Special quarterly exhibits help to produce a lively atmosphere, and popular lectures on scientific subjects connected with the museum objects are well attended.

Library Resources in the West Midlands

THE West Midlands Group of the Reference and Special Libraries Section of the Library Association has compiled a "Guide to Library Resources in the West Midlands" (pp. viii+49. London: Library Association, 1958. 15*s*.). Edited by B. G. Staples, it indicates the character, coverage and availability of 178 libraries in Herefordshire, Shropshire, Staffordshire, Warwickshire and Worcestershire. The staff, book-stock and number of periodicals, but not the titles, are given and only to a limited extent does it supply a picture of the resources of the region. There are two indexes; one, topographical, showing the different types of library in each locality; while the general index gives a key to particular subject fields and named collections, as well as indicating where microcard and microfilm readers and certain classes of material can be found. This well-produced pamphlet bears further witness to the preparatory work which this and similar groups are carrying out in different parts of Britain and on which an adequate regional library system may perhaps ultimately be based.

Health Physics

A NEW journal is a mixed blessing. The hopeful scribe sees in it another vehicle for his works, while the harassed executive sighs at the thought of more to read. Nevertheless, no one can grudge a new discipline its own organ. Health physics is such a new discipline; a Health Physics Society has recently been formed in the United States and has now published the first issue of its journal, *Health Physics* (1, No. 1, June 1958. Pp. 95. Subscription: 17 dollars; 120*s*. per annum. New York and London: Pergamon Press, 1958). In the foreword by K. Z. Morgan (editor-in-chief) health physics is defined "as a study and practice dealing with any and all factors relating to damage from ionizing radiation and the prevention of such damage". With the rapid promotion of nuclear energy and the increasing utilization of its by-products, there can be no doubt that the fantastic growth of this new corps of specialists from a handful of dedicated enthusiasts to a substantial army will be continued. They will welcome this journal devoted to "(1) Research, (2) Engineering, (3) Applied, and (4) General"—which is a fairly broad programme—and the first number, of June, has plenty of category (4).

"Atom 1958"

UNDER the title "Atom 1958" (pp. 24. London: H.M. Stationery Office, 1958), the Atomic Energy Authority has issued for general reading a summary, with illustrations, of the Authority's fourth annual report (see also p. 615 of this issue of *Nature*). The pamphlet describes in non-technical terms the progress of the power programme, the possibilities for the next two decades, including the prospects for a fusion reactor, and the relations of the Authority with industry and with other countries. Besides some notes on health and safety and the supply of fuel for reactors, it includes an outline of the Authority's organization and lists of its establishments and publications. It is a well-planned contribution to public relations which should help to remove some popular misunderstandings and dispel the dangerous sense of mystery with which those relations are sometimes cloaked.

Czechoslovak–Hungarian Polarographic Conference

THE Czechoslovak–Hungarian Polarographic Conference in Prague was held during July 13–24 following the similar meeting held in 1955 at the Hungarian Chemical University in Veszprém. The Conference was arranged by the Polarographic Institute of the Czechoslovak Academy of Sciences under the auspices of its Chemical Section. Prof. J. Heyrovský presided. The Hungarian delegation consisted of nineteen scientists and was led by Prof. J. Proszt (Budapest Technical University), Prof. K. Polinszky (dean, Veszprém Chemical University) and Dr. V. Cieleszky (chairman, Polarographic Section, Hungarian Chemical Society). Besides these, there were thirty-five visitors from Budapest and several polarographic experts from other countries, including Prof. G. Semerano (Padua); Dr. Z. Zagórski (Poznań); W. Stolle (German Academy, Berlin), who brought a gift of a newly constructed pen-recording polarograph; L. Lukashev (Bulgaria); Prof. B. P. Bruns (Moscow); and Dr. R. J. Magee (The Queen's University, Belfast). Sixty-three papers were presented at the Conference in two sections: *A*, inorganic–analytical; and *B*, organic and biochemical. After the meetings in Prague the delegates went to Bratislava and were shown around research establishments and chemical works in Czechoslovakia, as well as the Tatra Mountains. The majority of the papers were of high standard and some indicated new directions in polarography. They will be published chiefly in "Collection of Czechoslovak Chemical Communications". Those who are interested in the abstracts of the Communications should apply to Prof. J. Heyrovský, Polarographic Institute of the Czechoslovak Academy of Sciences, Vlašská 9, Prague 1, Czechoslovakia.

A Submarine Volcano

As reported by Y. Y. Gakkel (*Priroda*, 4, 87; 1958), the members of the polar drifting station in November 1955 discovered a submarine volcano situated at 88° 16′ N., 65° 36′ W. on the Lomonosov submarine ridge near the North Pole. The nature of the ice-quakes, noises and hydrogen sulphide emerging from a crack in the ice—all these phenomena have

suggested the presence of a submarine volcano in eruption. The author of this article postulates a tectonic fracture stretching along an arc from the Ellersmere Land to the Taimyr peninsula. Along this line the following newly discovered features can be located: the problematic volcano, dredged volcanic glass and basaltic hornblende, the latter being also found in other localities of the Arctic Ocean.

Seed Transmission of Plant Virus Diseases

Of the considerable number of known plant virus diseases, only some forty-five are reported to be seed-transmitted, and of these only a few cases of actual embryo infection are known. N. C. Crowley, who has already demonstrated the presence of virus inhibitors in the seeds of several species, but not of virus inactivators, has now made further contributions on this topic (*Austral. J. Biol. Sci.*, 10, 443 and 449; 1957). He has shown, by growing tomato embryos in media containing the virus, that there is no evidence for the presence of virus-inactivating substances in the embryos. Evidence is presented that whereas some of the highly infectious viruses commonly infect the seed testa, and even the endosperm, the developing embryo usually, and typically, remains uninfected. Since it has not been possible to obtain evidence of the presence of virus-inactivating agents in either the endosperm or the embryo, the favoured hypothesis to account for the rarity of seed-transmission is that the absence of plasmodesmatal connexions between the embryo and its tissue environment prevents its becoming infected. In a series of critical experiments, using well-known viruses and hosts, Crowley has shown to what extent the testa, the endosperm and the embryo may become infected in different cases. Again, all the evidence obtained indicates that the absence of seed-transmission of highly infectious viruses cannot be explained except in terms of the plasmodesmatal hypothesis. The alternative hypotheses (1) that the viruses induce sterility, (2) that the hosts contain inactivating substances, and (3) that developing embryos inactivate virus particles in the surrounding medium, are not validated by any evidence so far obtained. In the case of those virus diseases, for example, bean mosaic virus, that are seed-transmitted, this appears to depend on the ability of the virus to infect the microspores, macrospores or embryo sac before fertilization; but embryos themselves seem to be singularly free from infection. As the author points out, since natural selection would tend to eliminate any line of plants readily subject to the seed-transmission of a severe virus disease, the rarity of the latter phenomenon finds a simple and direct explanation.

Medical Research Council Travelling Awards

The Medical Research Council has made the following travelling awards for the academic year 1958–59. *Rockefeller Travelling Fellowships in Medicine:* Dr. G. Burnstock, Department of Pharmacology, Oxford; Mr. P. J. Holloway, Medical Research Council Laboratories, Hampstead; Mr. W. B. Jennett, lecturer in neurosurgery, University of Manchester; Dr. A. P. Mathias, lecturer in biochemistry, University College, London; Dr. R. P. Michael, Department of Experimental Neuroendocrinology, Institute of Psychiatry, Maudsley Hospital, London; Dr. P. H. A. Sneath, National Institute for Medical Research, Mill Hill. *Lederle Travelling Fellowship in Medicine:* Dr. R. J. Linden, lecturer in physiology, University of Leeds. *Alexander Pigott Wernher Memorial Travelling Fellowship in Ophthalmology and Otology:* Dr. G. B. Arden, Institute of Ophthalmology, London. *French Exchange Scholarships in Medical Science:* Miss J. E. Cremer, Medical Research Council Toxicology Research Unit, Carshalton, Surrey; Miss S. Reiman, Institute of Psychiatry, Maudsley Hospital, London.

Travelling Fellowships in Medicine have been awarded by the Eli Lilly Company of Indianapolis, after nomination by the Medical Research Council, to the following candidates: Mr. J. D. Griffiths, St. Bartholomew's Hospital, London; Dr. C. W. M. Wilson, lecturer in pharmacology and therapeutics, University of Liverpool. Travelling Fellowships in Medical Science have also been awarded by the United States Public Health Services, National Institutes of Health, Bethesda, Maryland, after nomination by the Medical Research Council, to the following candidates: Dr. B. S. Hartley, Department of Biochemistry, University of Cambridge; Dr. K. W. Cross, St. Mary's Hospital Medical School, London; Dr. A. Doig, Royal Infirmary, Edinburgh.

Announcements

The Ministry of Supply announces that Dr. J. S. McPetrie has been appointed director-general of Electronics Research and Development in succession to Dr. D. H. Black.

A meeting of the Photobiology Group on "Some Aspects of the Physiology of Vision" is to be held in the University Laboratory of Physiology, Oxford, during September 26–27. Registration forms and further details can be obtained from the chairman of the Photobiology Group, Dr. M. H. Pirenne, University Laboratory of Physiology, Oxford.

The Radio Frequency Spectroscopy Group (chairman, Prof. E. R. Andrew, Bangor) is to hold a conference on "Applications of Magnetic Resonance to Solid State Physics" at King's College, Newcastle upon Tyne (University of Durham), during September 22–23. The programme includes contributions on electron resonance of colour centres and iron group impurities in ionic crystals, nuclear resonance in ionic solids, electron spin, nuclear and cyclotron resonance in semi-conductors. Further particulars and application forms can be obtained from Dr. E. E. Schneider, King's College, Newcastle upon Tyne.

An international conference on "Small Angle X-ray Scattering from Metals" will be held at Midwest Research Institute, Kansas City, Missouri, during September 23–25. The purpose of the meeting is to bring together active workers in the field to discuss the origin of the scattering as it relates to the crystal structure of metals. Arrangements for the conference have been made by the following committee: C. S. Barrett, Institute for Study of Metals, University of Chicago; Paul A. Beck, University of Illinois; W. W. Beeman, University of Wisconsin; and J. C. Grosskreutz (chairman), Midwest Research Institute, Kansas City, Missouri, from whom further information can be obtained. Financial support is being provided by the National Science Foundation, the Office of Ordnance Research, the Office of Naval Research, the Wright Air Development Center, U.S. Air Force, and Midwest Research Institute. The proceedings will be published as a special section of the *Journal of Applied Physics*.

THE ACCIDENT AT WINDSCALE ON OCTOBER 10, 1957

THE final report* on the accident to No. 1 pile at Windscale on October 10, 1957, should be considered in relation to the previous reports[1–3].

During a routine Wigner energy release, temperatures in the pile rose beyond safe limits, causing a fire which affected 150 channels and caused a certain amount of radioactivity, mainly iodine, to be released over an area of Cumberland. Following this incident, a committee of inquiry was set up under Sir William Penney, and reporting to the Prime Minister. A less technical version of its report has already been published[1]. In the memorandum by the Prime Minister the immediate cause of the accident was given as "the application too soon and at too rapid a rate of a second nuclear heating to release the Wigner energy from the graphite . . .". The accident was considered as being due partly to inadequacies in the instrumentation and partly to faults of judgment by the operating staff, these faults of judgment being themselves attributed to weaknesses of organization.

Three committees were set up, each under the chairmanship of Sir Alexander Fleck, to advise on certain aspects of the incident. The committees on Organization and on Health and Safety reported in December 1957[2] and January 1958[3], respectively. The report of the Technical Evaluation Committee, the subject of this article, was delayed due to the need to carry out a considerable amount of experimental and analytical work.

The Committee's terms of reference were: (1) to consider the new information available following the accident; (2) to consider the possible methods of carrying out the controlled release of Wigner energy; and (3) to advise the United Kingdom Atomic Energy Authority on the restarting of Windscale No. 2 pile.

The report begins by considering the phenomenon of energy storage in irradiated graphite—Wigner energy. This storage is due to defects in the crystal structure of the graphite following neutron bombardment. The energy is releasable, in varying extent and rate, by heating the graphite. The amount of energy stored decreases rapidly with an increase in the temperature at which irradiation takes place, that is, there is an annealing effect. This feature makes the effect particularly severe in low-temperature reactors such as BEPO and the Windscale piles, but very much less important in the Calder and subsequent power-producing reactors. The Committee states that if the release of energy in No. 2 pile was attempted as previously, graphite temperatures in excess of 400° C. would almost certainly occur in considerable masses of graphite, and that temperatures in some pockets where previous releases had not been effective might reach 500° C.

The report next considers the implications of these transient high temperatures on the graphite, the fuel and the other materials in the reactor. A temperature limit of 400° C. is considered as reasonable for the graphite. Under these conditions the fuel element design is satisfactory. Having specified temperature limitations, the real problem is that of installing sufficient and effective instrumentation. A considerable increase in the number of graphite and fuel element thermocouples is required, and the fission product detection scanning gear needs improving.

A description of four methods of carrying out Wigner releases is then given. The first method, the one previously employed, uses nuclear heating to initiate the release of energy. The main difficulty is the non-uniformity in the spread of the release, leaving pockets unannealed. The Committee deprecates the application of a second nuclear heating to initiate the release of these pockets, as being certain to lead to excessive uranium temperatures, and recommends that no further releases by this method should be attempted. The second method was used with the BEPO reactor in March this year, but is not recommended for Windscale. The third method, a slow heating method, is also discarded, since it would cause excessive stresses in the concrete reactor structure. The fourth method combines the previous two. It is proposed that a partial anneal using a slow heating method should first be carried out, followed by a more rapid heating to a somewhat higher temperature. The heat would be supplied by heating the ingoing air. The necessary plant and instrumentation could be installed in 6–9 months, making the pile operational again in about a year.

No. 1 pile has added considerably to our knowledge of reactor technology and operation. It is hoped that No. 2 pile will be recommissioned, since the development of methods for altering the pattern of the instrumentation in a reactor after it has been in service will prove most useful as the nuclear power programme develops.

* Atomic Energy Office. Final Report of the Committee appointed by the Prime Minister to make a technical evaluation of information relating to the design and operation of the Windscale piles, and to review the factors involved in the controlled release of Wigner energy. Pp. 20. (Cmnd. 471.) (London: H.M. Stationery Office, 1958.) 1s. net.

[1] Accident at Windscale No. 1 pile on 10th October, 1957. (Cmnd. 302.) (H.M. Stationery Office.) *Nature*, **180**, 1043 (1957).

[2] The organization of certain parts of the U.K.A.E.A. (Cmnd. 338.) (H.M. Stationery Office.) *Nature*, **181**, 439 (1958.)

[3] The organization for control of health and safety in the U.K.A.E.A. (Cmnd. 342.) (H.M. Stationery Office.) *Nature*, **181**, 1023 (1958).

EXCHANGE OF INFORMATION ON ATOMIC ENERGY

THE Agreement between the Governments of the United Kingdom and of the United States for Co-operation on the Uses of Atomic Energy for Mutual Defence Purposes described in the White Paper* published on July 8 comprises twelve articles. The first is a general provision for the exchange of information and transfer of materials and equipment, unless such co-operation represents an unreasonable risk to the defence and security of the communicating or transferring party. Under the second article the two parties agree to communicate or exchange classified information jointly determined as necessary to the development of defence plans, the training of personnel in the use of or in defence

* Agreement between the Government of the United Kingdom of Great Britain and Northern Ireland and the Government of the United States of America for Co-operation on the Uses of Atomic Energy for Mutual Defence Purposes, Washington, July 3, 1958. Pp. 8. (Cmnd. 470.) (London: H.M. Stationery Office, 1958.) 6d. net.

against nuclear weapons, the development of delivery systems for such weapons, and on research, development and design of military reactors, as well as other classified information required to improve the design, development and manufacture of nuclear weapons. The transfer of submarine nuclear propulsion plant and materials is covered specifically in the fourth article, while questions of responsibility, conditions and guarantees are dealt within the next three. Article VII restricts the communication of classified information or material transferred under the Agreement to third parties without prior authorization, and under Article VIII agreed classification policies are to be maintained and the present practice of consultation continued. The longest article (IX) is that dealing with patents, and includes provision for the issue of appropriate secrecy or prohibition orders. These clauses, however, make it possible for firms in one country to enter directly into relations with firms in the other on the sale of patents and licences, and should thus ease the exchange of information between the two countries where it had been hindered because information which was the property of individual British firms would, if exchanged, become the property of their American competitors.

As the Prime Minister explained in the House of Commons on July 8, the new Agreement, which supersedes that of June 1955, has been negotiated in consequence of the amendment of the United States Atomic Energy Act, 1954, so as to permit closer and more fruitful collaboration between Great Britain, the United States and other friendly countries. The Agreement was signed on July 3 but cannot become effective until it has lain before Congress for thirty sitting days, and the Prime Minister preferred not to comment at that time on the practical results which might be expected when the Agreement comes into effect. We had been in close touch with the Commonwealth countries during the negotiations leading to the Agreement and co-operation with these countries was covered by Article VII. On July 15 Mr. Macmillan added that the United States Atomic Energy Act, as amended, still did not permit the transfer of complete nuclear weapons or manufactured nuclear components of such weapons, and the Agreement accordingly dealt rather with research and development than with the actual interchange of weapons. Replying to further questions in written answers on July 17, Mr. R. A. Butler said that no commitments or understandings were entered into which might have the effect of surrendering patent rights in the Calder Hall type of reactor. British interests were satisfactorily safeguarded by the clauses of the Agreement, and the Government saw no reason to expect that the undertakings to make available to the United States Atomic Energy Commission information about the Calder Hall design and the standard fuel elements used in the Calder Hall reactors would have any substantial effect on our export prospects. In the House of Lords on July 21, the Earl of Gosford said that this information related only to the Authority's own dual-purpose reactors and did not and would not include information developed from Calder Hall and included by industry in its latest nuclear power station designs. The Agreement was concerned exclusively with co-operation in the use of nuclear energy for defence and not with arrangements for co-operation on civil uses.

COLONIAL DEVELOPMENT

THE schemes made under the Colonial Development and Welfare Acts in the period April 1, 1957–March 31, 1958, which are detailed in the latest return*, bring the total commitments, from April 1, 1946, under the Acts to £194,259,668, of which £17,203,311 is for research schemes. The totals for the year in question are £17,480,563 for development and welfare and £1,123,850 for research schemes. Of this last, £351,709 is for agriculture, £158,830 for tsetse and trypanosomiasis, £106,207 for medicine, £101,108 for insecticides, £76,859 for fisheries, £72,622 for social science, £52,240 for products research, £23,546 for locust control, and £14,340 for economic research.

Of the actual research schemes, £135,730 is supplementary provision for the East African Trypanosomiasis Research Organization, 1957–60; £70,156 for the Colonial Pesticides Research Unit, Arusha, 1957–58; £66,858 for maintenance of the Colonial Road Research Section at the Road Research Laboratory, Harmondsworth; £65,000 for expansion of vaccine production and associated research in Nigeria; £51,675 for agricultural research in Swaziland; a further £49,300 for expansion of the Tropical Products Institute; £49,064 for the East African Scientific and Industrial Research Organization, 1957–60; a supplementary £43,056 for the Colonial Microbiological Research Institute, Trinidad; £37,785 for soil and land use surveys in British Guiana; £34,185 to establish a Veterinary Investigational Laboratory in Bechuanaland; £32,992 for the Nigerian Institute of Social and Economic Research; and £30,938 for the joint capital housing programme for the East African Agricultural and Forestry Research and Veterinary Research Organizations.

Other major research schemes include £23,546 for the Desert Locust Survey; a supplementary £30,800 to establish a Soils Laboratory in Sarawak; £23,023 for the study of soil fertility maintenance in high-rainfall areas in Nigeria; a further £22,084 for the Fisheries Development and Research Station, Sierra Leone; a further £20,100 for the West African Institute for Trypanosomiasis Research; £19,370 for control of malaria in a hyperendemic area of East Africa; and a supplementary £15,833 for sugar technology research at the College of Tropical Agriculture, Trinidad.

The development and welfare schemes follow the general pattern of recent years. A further grant of £644,000 goes to the Central Organization for Overseas Geodetic and Topographical Survey; Kenya receives £507,880 for the intensification of African agricultural development, with a further £508,120 for Agricultural Department expenditure in connexion with that intensification. Farm development in Jamaica receives a further £750,880, with £177,375

* Colonial Development and Welfare Acts. Return of Schemes made under the Colonial Development and Welfare Acts by the Secretary of State for the Colonies, with the concurrence of the Treasury, in the period from 1st April, 1957, to 31st March, 1958. Pp. 36. (House of Commons Paper No. 223.) (London: H.M. Stationery Office, 1958.) 2s. net.

for expansion of cocoa production. £156,973 goes for soil conservation in Basutoland; a further £81,394 for control of tsetse fly in the Bechuanaland Protectorate; and £234,000 for development of broadcasting services in Kenya. Among the many grants for road development may be mentioned £771,680 to the Western Region of Nigeria; £288,000 to Tanganyika (a further £280,000 for improvements to the Eastern Trunk Road); £300,000 for improvement of territorial roads in Kenya; £156,000 for survey of roads in Swaziland; £817,250 for construction of a trunk road from Scrian to Simanggang, Sarawak; and £103,692 for reconstruction of the East Coast Georgetown–Rosignol road, British Guiana. Grants of £464,295 to the Western Region, Nigeria, and £99,000 for the Scaries area, Sierra Leone, illustrate the grants for the development of water supplies. £200,000 is for the establishment of the Jamaica Institute of Technology, and among the many grants for university education can only be mentioned £198,000 for the erection of a Men's Hall of Residence at the University College of East Africa and a supplementary £154,650 for the erection of the new library; £200,000 towards a new library at the University of Hong Kong; and £156,188 for the construction of buildings for the Faculty of Medicine, University College of the West Indies. There are numerous grants for secondary and primary education, the largest being £584,000 for the expansion of general education in Lagos.

SAFETY IN MINES IN GREAT BRITAIN

THE Mines Inspectorate, apart from the Headquarters organization at London, is divided into eight divisions, namely, Scottish, Northumberland and Cumberland, Durham, North Eastern, North Western, East Midland, West Midland and Southern, and South Western. Each division is in the charge of a divisional inspector who is required, under the Coal Mines Act of 1911, to present an annual report to the Minister of Power. The reports* for each of the eight divisions for 1956 have been published at intervals during the first few months of 1958; these are the last reports under the 1911 Act, for the Mines and Quarries Act, 1954, became effective on January 1, 1957.

The reports follow a similar pattern, beginning with an introduction which contains short accounts of production in the divisions, reorganization of collieries, new work, introduction of new methods of working, new machines used and experimental work on techniques of mining, roof support, shot firing, ventilation and so forth. This is followed by a section devoted to administration, in which are summarized the establishment and organization of the staff, the activities of the inspectors, inspections made on behalf of workmen, and prosecutions. One of the most interesting and significant features of this section in the reports is that nearly every divisional inspector refers to a serious shortage of staff, although one or two gave their opinion that there were signs of improvement in the near future. The shortage of inspectors has meant that the amount of preventive work undertaken was much less than desirable. The effect of the shortage was aggravated by the additional work caused in preparation for the implementation of the 1954 Act at the beginning of 1957; for example, in one division alone the Divisional Committee of the Mining Qualifications Board (a committee which includes inspectors of mines) examined the qualifications of more than 2,000 mechanics and examined about 100 of these orally; and certificates were issued in that division for more than 500 deputies and for nearly 900 shot-firers.

* Ministry of Power. Reports of H.M. Inspectors of Mines under the Coal Mines Act 1911, for 1956. Durham Division. By W. Brown. Pp. ii+30+8 plates. 2s. 6d. net. North Western Division. By G. Hoyle. Pp. ii+36+4 plates. 3s. net. North Eastern Division. By C. W. Scott. Pp. ii+38+6 plates. 2s. 6d. net. Scottish Division. By W. Widdas. Pp. ii+28+4 plates. 2s. 6d. net. East Midland Division. By W. B. Brown. Pp. ii+34+4 plates. 3s. net. South Western Division. By T. A. Jones. Pp. ii+24+4 plates. 2s. 6d. net. West Midland and Southern Division. By J. E. Henshaw. Pp. ii+24+2 plates. 2s. 6d. net. Northumberland and Cumberland Division. By H. Hyde. Pp. ii+30+6 plates. 2s. 6d. net. (London: H.M. Stationery Office, 1958.)

Over Great Britain as a whole, proceedings were taken against two management officials. Managements took legal proceedings against subordinate officials and workmen in about 340 cases, more than half of which were for contraventions in connexion with contraband (matches and smoking); two-thirds of these cases occurred in one division.

The main section of each report deals with accidents and their prevention. During the year there were 300 fatal accidents at mines (compared with an average of 396 in the three preceding years), and more than 1,760 men reportably injured (about 1,900 on the average in the three preceding years). There were also 148 dangerous occurrences (gas ignitions, fires, breakages of winding gear and so forth). Accidents are investigated and analysed by the inspectors. There is, considering the country as a whole, a gradual downward trend in the number of fatalities and reportable injuries each year, and in two divisions they were the lowest ever recorded; the numbers are, as the inspectors stress, unduly high. An analysis of all the accidents in 1956 shows that, in the opinion of the examining inspectors, about 80 per cent were avoidable (the actual percentages in different divisions vary from 76·3 to 81·7) and the remaining 20 per cent "unavoidable under existing conditions".

Of the avoidable accidents, some were due to defective machinery, some to neglect or breach of rules by officials or by workmen, but most (from about a half to three-quarters of avoidable accidents in different divisions) were classified as "avoidable by ordinary caution". Over Great Britain as a whole, about 35 per cent of the accidents were due to falls of ground, about 25 per cent were on haulage and transport, about 30 per cent due to miscellaneous causes, and about 10 per cent were due to accidents on the surface.

Sections in each of the reports are devoted to safety organization, training and education, and health and welfare. A noteworthy feature is the interest taken by miners in rescue training and rescue competitions, in first-aid work and competitions and in civil defence work. The rescue organization of the coal mining industry, which depends largely upon rescue teams of volunteers trained at special rescue stations, is a fine one and, as reported by the inspectors, whose duties include inspection of the rescue stations, there is no lack of volunteers for rescue work.

The inspectors, as their reports show, take a keen interest in training and education, and report on the training centres, training faces and courses of instruction in each division. In 1956 more than 17,000 juveniles and more than 15,000 adults received preliminary training; these figures indicate the immense task of training in the coal industry, especially as face training has to be arranged and courses for potential shot-firers and potential deputies (quite apart from courses for senior officials, which are not referred to in detail).

Sections on health and welfare refer mainly to work carried out in improving dust control, ventilation, lighting, sanitation and so forth, and to the provision of medical centres at collieries.

Each divisional report includes a section devoted to horses. Although the number of horses employed at collieries is decreasing, more than 10,000 were employed in 1956. In general, the miner is devoted to his horses and they are well cared for.

Although the general pattern of the reports is similar, they are not stereotyped. Probably few people unconnected with the mining industry will read them, but they are very well worth reading, for they are personal reports and are far from the dry-as-dust documents they might have been.

The reports for 1957 of H.M. Inspectors of Mines and Quarries under the Mines and Quarries Act, 1954, have also recently been published; these are the first reports under the 1954 Act which became effective on January 1, 1957. It makes provision for all classes of mines and quarries to be dealt with together, and the Inspectors' reports include, in separate parts or sections, reports on coal mines; mines of stratified ironstone, shale and fireclay; miscellaneous mines (including metalliferous mines); and quarries*.

The general pattern is similar to the earlier divisional reports, which dealt only with coal mines and other so-called stratified mines. As may well have been expected, by far the greater part of each report deals with coal mines and the conclusions are similar to those for 1956.

Until 1957 the reports of H.M. Inspectors of Mines and Quarries on metalliferous mines and quarries were presented in a single publication for the whole country by H.M. Chief Inspector of Mines and Quarries under the Metalliferous Mines Regulation Act, 1872, and the Quarries Act, 1894; the last report under these Acts was published early in 1958 and referred to the years 1954–56. The new arrangement under the 1954 Act whereby H.M. Inspectors report annually on all mines and quarries in their division is more convenient in many ways than the former biennial reports.

The new reports show that particular attention is being paid at quarries to dust prevention and dust suppression and to encouragement in the use of protective clothing, especially safety hats, which many quarrymen seem loath to wear. S. G. Ward

* Reports of H.M. Inspectors of Mines and Quarries under the Mines and Quarries Act, 1954, for 1957. Northumberland and Cumberland Division. By H. Hyde. Pp. iv+41+6 plates. 3*s*. 6*d*. net. Scottish Division. By W. Widdas. Pp. iv+41+6 plates. 4*s*. net. North Western Division. By G. Hoyle. Pp. ii+42+6 plates. 3*s*. 6*d*. net. Durham Division. By W. Brown. Pp. iv+23. 2*s*. 6*d*. net. South Western Division. By T. A. Jones. Pp. iv+28+4 plates. 2*s*. 6*d*. net. West Midland and Southern Division. By J. E. Henshaw. Pp. iv+34+2 plates. 3*s*. net. North Eastern Division. By C. W. Scott. Pp. ii+44+7 plates. 3*s*. 6*d*. net. East Midland Division. By W. B. Brown. Pp. ii+38+7 plates. 3*s*. 6*d*. net. (London: H.M. Stationery Office, 1958.)

PRESSURE WELDING OF METALS

WELDING engineering is a profession now well established in industry and is supported by a research association and a technical institute. In Britain (unlike the United States and the U.S.S.R.) it has not achieved independent status at any university, and it is therefore practised by metallurgists and mechanical, electrical and civil engineers, in whose training it has occupied a very subsidiary role. However, as a result of the division of metallurgy into physical and industrial aspects at the University of Birmingham, research into and the teaching of the basis of metallic jointing practice now figures prominently in the Department of Industrial Metallurgy. To supplement the academic work, occasional conferences are organized to promote the exchange of information and ideas with other workers in the field. The second of these conferences, on the pressure welding of metals, was held on June 19, under the chairmanship of Prof. E. C. Rollason, head of the Department of Industrial Metallurgy, and was attended by ninety representatives of industry, the research associations and the universities.

After explaining the purpose of these conferences, Prof. Rollason reviewed the origins and development of the practice of pressure welding and the progress that had been made towards establishing a theoretical model. Three papers were then presented, the first by Mr. J. A. Donelan (Research Laboratories, General Electric Co.) dealt with industrial practice, the second by Messrs. L. R. Vaidyanath, M. G. Nicholas and D. R. Milner gave an account of the researches being carried out at the University of Birmingham, and the third paper by Dr. E. Holmes (University of Nottingham) was concerned with the relative importance of movement at the interface, oxide break-up, and the deformation of surface asperities upon bond formation.

The idea of joining metals by pressure is not new, as it has been employed in the hammer-forged blacksmith's weld from the earliest days of the use of iron; this, however, is a special case in that it depends on the squeezing out of a molten layer of slag between the two components to be joined. The general practice of pressure welding metals, without melting, was established in 1887 with resistance heating used to aid the deformation of the interface; then, during the Second World War, the cold welding process was developed in which metals are joined at room temperature. The majority of metals can now be pressure welded, if not at room temperature, then at higher temperatures. The advantages of the process are that the joint possesses none of the weaknesses inherent in the cast structure associated with the more normal welding techniques, and it is the only metallic jointing process that can be carried out at room temperature, and is therefore particularly suitable to the canning of delicate components such as detonators or transistors. One of the widest applications of pressure welding is to the production of ply metals by rolling, so that the advantages of the properties of two or more metals or alloys can

be combined, for example, to provide corrosion resistance, as in the cladding of high-strength aluminium alloys with a thin layer of super-purity aluminium, or for economical reasons as in the cladding of mild steel with a thin layer of stainless steel.

Pressure welding practices developed empirically, and it was soon found that metals varied markedly in their welding characteristics, the criterion of weldability usually adopted being the amount by which two pieces of metal when placed together have to be deformed in order for them to join : thus, for example, at room temperature overlapped strips of tin and lead require a reduction in thickness of about 20 per cent, aluminium and copper 50 per cent, and nickel and iron 80 per cent. Cold welding is largely applied to aluminium and copper, and Mr. Donelan cited many applications in the electrical industry such as joining the ends of wires and rod, the formation of cable sheath from strip, and the fabrication of electric kettles and refrigerator coolant panels. Mr. Donelan stressed that for successful welding the most important factors are the surface preparation of the material (a degreasing treatment followed by scratch brushing being preferred), and the shape of the deforming tool.

The understanding of the phenomenon of pressure welding lags behind its practice and no satisfactory theory has been developed, although several hypotheses have been advanced as to the nature of the controlling parameter. When pressure welds could only be made at high temperatures it was considered that diffusion at the interface was essential to bring the surfaces into atomic contact, and for this reason that welding was enhanced at the recrystallization and transition temperatures, at which the rate of diffusion was considerably increased. With the advent of cold pressure welding emphasis was transferred from the influence of the temperature of welding to the pressure applied, which then came to be regarded by some workers as the controlling factor. These hypotheses, however, take little account of the practical necessity for careful surface preparation before welding, and more recent theories have concentrated on this aspect, in one case the penetration and fragmentation of the oxide film being regarded as critical, whereas in another case the difficulty of bringing roughened surfaces into intimate contact has been stressed. An alternative hypothesis has been advanced which implicitly assumes that the formation of bonds presents no difficulty, but that on release of the applied load these bonds are at least partially broken apart by elastic recovery forces which vary from one metal to another. Finally, there is the possibility of relative movement between the joining surfaces which, as workers in the field of friction have shown, leads to the formation of welds that are responsible for frictional restraint. None of the models suggested has been developed in detail and none of them can explain all the observed facts.

The difficulties in making an analysis of bond formation are : first, the large number of parameters involved ; and second, the fact that no satisfactory method has been developed for the measurement of the degree of bonding or weld strength which is obtained. Part of the theme of the work of Vaidyanath, Nicholas and Milner was that the mechanism of pressure welding is best investigated via the roll bonding process, since for practical purposes the reduction is uniform throughout the composite and it is possible to measure the bond strength by a test giving results which are more amenable to analysis than those available hitherto. This work has led to the development of a theoretical model for the maximum joint strength that can be obtained from the concept that oxide films are completely brittle, so that when the interface is extended, areas of virgin metal are exposed and these are the sources of potential bonding. However, the problem remains of why these virgin metal areas do not always join. From evidence on the effect of pressure, time, temperature, and exposure of the surface to the atmosphere prior to rolling, the authors concluded that a correct theory must involve considerations of atomic movements during deformation, diffusion and the solution of entrapped gases.

When dissimilar metals are welded, further problems arise in that if the two metals possess different mechanical properties they will not deform in the same manner and, in addition, alloy layers will develop at the interface and have a marked effect on the joint strength. Dr. Holmes showed that when dissimilar metals are bonded by rolling there will also be relative movement between the two surfaces as they enter the roll gap and that this could lead to substantial bonding without much deformation, the bond strength increasing with the amount of relative movement and with the applied load under which the movement was carried out. From further experiments, designed to examine the relative importance of the difficulty of bringing rough surfaces into intimate contact and that of penetrating the oxide film, Dr. Holmes reached the conclusion that for conditions under which the flow of metal was restrained oxide break-up was difficult to obtain, whereas, since restraint of necessity involves higher pressures, the deformation of asperities was readily achieved. Conversely, for conditions of low restraint, metal flow and oxide break-up took place readily and the deformation of the surface asperities became the controlling process.

The points raised during the discussion showed that pressure welding is contributing to the solution of some of the problems arising from modern technical developments, two examples of the use of cold welding being the joining of the trans-Atlantic cable and the fabrication of complex patterns of frozen mercury for precision casting. The disadvantage remains that in many cases considerable deformation is required, but a recent development has shown that this can be very much reduced by the application of ultrasonic vibration during the welding process. The use of impact loading was also discussed, but the results were inconclusive ; some indicated little difference, while others suggested considerable improvements, from the bond strengths obtained by deformation at conventional speeds. The divergence of the opinions expressed on the theory of pressure welding showed how little can be regarded as universally acceptable, and the ultimate answer will probably show that the controlling parameter varies with the conditions of welding. The technique of surface preparation prior to welding received considerable discussion, and it is found that the best welds are achieved with scratch-brushed surfaces : with many other surface treatments, or no surface preparation, it is often impossible to obtain any bonding. That no satisfactory explanation could be offered for the success of the scratch-brushing practice demonstrated again the inadequacy of the present state of the theoretical treatments of the subject.

D. R. Milner

USE OF ARTIFICIAL SATELLITES TO EXPLORE THE EARTH'S GRAVITATIONAL FIELD: RESULTS FROM *SPUTNIK* 2 (1957β)

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IT is well known that the Earth's oblateness and its atmosphere cause the main perturbations to the orbit of a near Earth-satellite, and various theoretical studies of these effects have been made. Now that accurate observations of satellite orbits are available it becomes possible, at least in principle, to reverse this procedure and to use the theories to obtain a better knowledge of the atmosphere and the Earth's gravitational field. This article gives some tentative conclusions about the gravitational field, after a comparison of theory with observation for *Sputnik* 2 (1957β).

Fortunately, it happens that the first-order perturbations due to the atmosphere and the Earth's oblateness are of different kinds: the air drag steadily reduces the length of the major axis and the eccentricity of the orbit[1], while the Earth's oblateness causes the orbital plane to rotate about the Earth's axis and the major axis of the orbit to rotate in the orbital plane. The rotation of the major axis is difficult to determine; but, as Buchar[2] has remarked, the rotation of the orbital plane, which can be measured very accurately, should provide new information about the gravitational field.

The gravitational potential U due to the Earth at an exterior point distant r from the centre may be expressed in spherical harmonics $S_n(\theta,\varphi)$ as:

$$U = \sum_{n=0}^{\infty} A_n r^{-n-1} S_n(\theta,\varphi) \qquad (1)$$

where θ is the angle between the radius vector and the Earth's axis (the co-latitude), φ is the longitude and the A_n are constants. If the potential is assumed to be independent of longitude and the equatorial plane is suitably chosen, equation (1) may be written more explicitly as:

$$U = \frac{fM}{R}\left\{\frac{R}{r} - \sum_{n=2}^{\infty} J_n\left(\frac{R}{r}\right)^{n+1} P_n(\cos\theta)\right\} \qquad (2)$$

where f is the gravitational constant, M the mass of the Earth, and R the Earth's equatorial radius. P_n is the Legendre polynomial of order n and the J_n are constants. Of the coefficients J_n, only J_2 is at present known at all accurately, and it is customary to express U in the form recommended by Jeffreys[3] as:

$$U = \frac{fM}{R}\left\{\frac{R}{r} + J\left(\frac{R}{r}\right)^3\left(\frac{1}{3} - \cos^2\theta\right) + \frac{D}{35}\left(\frac{R}{r}\right)^5(35\cos^4\theta - 30\cos^2\theta + 3)\right\} \qquad (3)$$

where the coefficients J and D are related to the J_n of equation (2) by $J = \frac{3}{2}J_2$ and $D = -\frac{35}{8}J_4$.

The geopotential Ψ is obtained by adding a term $\frac{1}{2}w^2r^2\sin^2\theta$ to U, to allow for the angular velocity w of the Earth, and Jeffreys chose the coefficient D in (3) to make the value of Ψ constant over a spheroid of equatorial radius R and polar radius $R(1-\varepsilon)$, which was taken as an approximation to the real Earth, with ε as the polar flattening. With these assumptions:

$$J = \varepsilon - \frac{m}{2} - \varepsilon\left(\frac{\varepsilon}{2} - \frac{m}{7}\right) + 0(\varepsilon^3) \qquad (4)$$

and

$$D = \frac{\varepsilon}{2}(7\varepsilon - 5m) + 0(\varepsilon^3) \qquad (5)$$

where $m = R^3w^2(1-\varepsilon)/fM$ is the ratio of centrifugal to gravitational acceleration at the equator, multiplied by $(1-\varepsilon)$. The value of m is accurately known ($10^6 m = 3449{\cdot}79 \pm 0{\cdot}06$), while from geodetic measurements Jeffreys deduced that $J = 0{\cdot}001637 \pm 0{\cdot}000004$, which gives $1/\varepsilon = 297{\cdot}1 \pm 0{\cdot}4$. Then from (5), $D = 10{\cdot}6 \times 10^{-6}$.

The mean rate of rotation of the orbital plane, $\dot{\Omega}$, for a near satellite in the gravitational field defined by (3) is found[4] to be:

$$\sqrt{\frac{fM}{a^3}}\left(\frac{R}{\bar{r}}\right)^2\cos\alpha\left[J + J^2\left(\frac{R}{\bar{r}}\right)^3\left(\frac{19}{12}\sin^2\alpha - 1\right) - \frac{3DR^2}{14\bar{r}^2}(7\sin^2\alpha - 4) + 0(J^3\varepsilon)\right] \qquad (6)$$

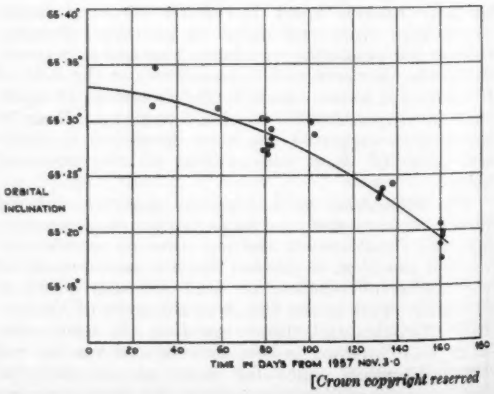

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Fig. 1. Mean orbital inclination of *Sputnik* 2 (1957β)

in the opposite direction to the satellite's motion, where a is the semi-major axis of the orbit, $\bar{r}$ the semi-latus rectum, e the orbital eccentricity and α the mean inclination of the orbital plane to the equator.

The expression (3) for U implies that the Earth is symmetrical about the equatorial plane. In considering the observational results, however, it is as well to remove this restrictive assumption and to include the third harmonic, a term :

$$\frac{fM}{R} J_3 \left(\frac{R}{\bar{r}}\right)^4 \tfrac{1}{2}(5\cos^3\theta - 3\cos\theta)$$

on the right-hand side of (3). It has been found (Merson, R. H., and Plimmer, R. N. A., unpublished work) that this gives rise to an extra term :

$$-\frac{3}{8}\sqrt{\frac{fM}{a^3}}\left(\frac{R}{\bar{r}}\right)^3 J_3 e \sin\omega \cot\alpha\,(15\sin^2\alpha - 4) \quad (7)$$

in (6), where ω is the mean argument of perigee.

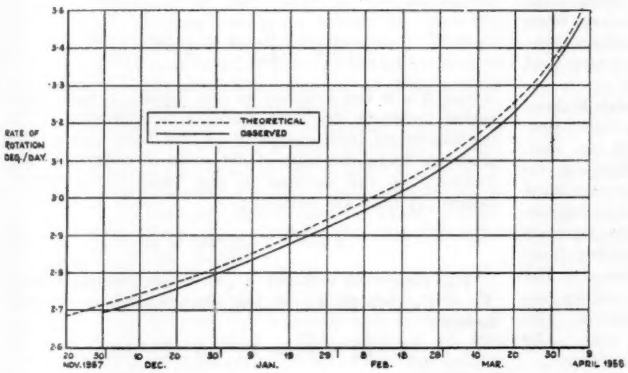

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Fig. 2. Rate of rotation of orbital plane of *Sputnik* 2

In the theoretical formula for $\dot{\Omega}$, as given by (6), the value of cos α is much less accurately known than the other factors outside the square brackets, and in comparing the theoretical and observed values of $\dot{\Omega}$, it is this uncertainty in cos α which sets the limit to the accuracy. The value of α for *Sputnik* 2 has been determined from a series of kinetheodolite observations made at Ministry of Supply stations at Aberporth, Bramshot, Larkhill, Orfordness and West Freugh. These have been analysed using a Pegasus digital computer at the Royal Aircraft Establishment (the method of analysis is described elsewhere[5,6]) and the results are plotted in Fig. 1. It is found that

$$\alpha = 65{\cdot}331° - 0{\cdot}00018°\,t - 0{\cdot}0000043°\,t^2 \pm 0{\cdot}02° \quad (8)$$

where t is the time in days after November 3·0, 1957.

This marked decrease in α, at the rate of about 0·001 deg./day, has not yet been satisfactorily explained. The theoretical formula for $\dot{\alpha}$ for an orbit *in vacuo* contains terms in J_3e, De^2 and J^2e. The value of $\dot{\alpha}$ given by this formula does not appear to be large enough to account for the observed change, however. Also, if the decrease were due to the J_3e term, the rate of decrease should tend to zero at the end of the life-time of the satellite when $e \to 0$, whereas Fig. 1 indicates that the decrease becomes sharper towards the end. This suggests that the change may be due to some second-order effect of the atmosphere, and possible causes, such as rotation of the atmosphere, are being investigated.

The theoretical value of $\dot{\Omega}$ during the life-time of *Sputnik* 2 has been calculated from the expression (6), using the values of J and D recommended by Jeffreys[3], values of α from (8), and values of other orbital elements derived from kinetheodolite observations and from theodolite observations made at Jokioinen, Finland. At the same time, observed values of Ω, the right ascension of the ascending node, have, after suitable curve-fitting, been differentiated and values of $\dot{\Omega}$ (observed) have been obtained. Fig. 2 shows the variation with time of the theoretical and observed values of Ω. It was found that the ratio $\dot{\Omega}$ (theoretical)/$\dot{\Omega}$ (observed) remained virtually constant during the life-time of *Sputnik* 2 and had the value $1{\cdot}0068 \pm 0{\cdot}0003$.

Since this ratio remained so nearly constant, it seems highly improbable that the discrepancy is due to atmospheric effects, which should increase steadily as time goes on. The effect of the J_3 term can also be discounted since the factor $e \sin\omega$ in (7) decreases markedly during the life-time of the satellite. A change in either J or D, however, would change $\dot{\Omega}$ by a nearly constant factor, because the ratio of the J-term to the D-term in (6) does not vary by more than 10 per cent during the life-time of the satellite. It therefore seems probable that one or both of the constants J and D is in need of revision.

The discrepancy between the observed and theoretical values of $\dot{\Omega}$ could be explained either by a change in J from 0·001637 to 0·001626 (corresponding to a change in $1/\varepsilon$ from 297·1 to $298{\cdot}1 \pm 0{\cdot}1$), or by a change in D from $10{\cdot}6 \times 10^{-6}$ to 45×10^{-6}. How the changes should be apportioned between J and D can best be decided by accurate observations on a satellite with different orbital inclination. The preliminary information so far available to us on the first of the three U.S. satellites, which had inclinations near 35°, suggests that the observed values of $\dot{\Omega}$ are, as for *Sputnik* 2, lower than the theoretical. Since the D term in (6) changes sign when α changes from 65° to 35°, it seems likely that the discrepancy is due mainly to the J-term and that J needs to be reduced, though not necessarily to the value given above.

We wish to thank the kinetheodolite observers of the Ministry of Supply, and Dr. G. Järnefelt, of the Jokioinen Meteorological Observatory, Finland, for providing the observations on which these results are based.

[1] King-Hele, D. G., and Leslie, D. C. M., *Nature*, **181**, 1761 (1958).
[2] Buchar, E., *Nature*, **182**, 198 (1958).
[3] Jeffreys, H., "The Earth", 3rd edit. (Camb. Univ. Press, 1952).
[4] King-Hele, D. G., *Proc. Roy. Soc.*, A (in the press).
[5] King-Hele, D. G., and Merson, R. H., *J. Brit. Interplan. Soc.* (August 1958).
[6] Merson, R. H., *Astronautica Acta* (August 1958).

AN APPARATUS FOR MICROELECTROPHORESIS OF SMALL PARTICLES

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Introduction

THE most widely used apparatus for electrophoresis of small particles have employed either rectangular[1–4] or cylindrical[5–9] chambers. The former has gained more adherents recently because of the optical difficulties[10,11] associated with cylindrical designs. The apparatus now described has been developed especially for studying the surface properties of tissue cells where rapid measurement and accurate temperature control are required.

Three features of this apparatus distinguish it from others previously used: (1) The tube is fixed permanently in position and is washed out *in situ*. (2) A dial test indicator is incorporated, allowing the microscope to be rapidly focused at the correct level independent of any inaccuracies in the fine adjustment screw. (3) The objective and electrophoresis tube are immersed in water, which simplifies temperature control and reduces the refraction by the cylindrical lens of the capillary wall. A reading can be taken and the tube cleaned in five minutes. The readings are reproducible to within 3 per cent. The apparatus is simple, robust and easy to construct.

Theory of the Cylindrical Cell

There are two electrokinetic effects in a closed glass tube containing a suspension of particles in an applied electric field. There is motion of the particles with respect to their suspending medium (electrophoretic effect) and a movement of liquid relative to the wall of the capillary (electro-osmotic effect) with a return flow of liquid down the centre of the tube. This produces a parabolic distribution of particle velocities across the diameter of the tube. The mobility of particles is measured at the stationary level, the position of which is derived below.

Lamb[12] has shown that for the closed cylindrical tube:

$$V_w = \frac{P}{4\eta}\,(r^2 - \tfrac{1}{2}R^2) \qquad (1)$$

where V_w is the velocity of the liquid, P the hydrostatic pressure gradient parallel to the axis, η the coefficient of bulk viscosity of the liquid, and r the distance from the axis of the tube of radius R. The velocity at the surface of the tube $V_s = PR^2/8\eta$; hence (1) may be written:

$$V_w = V_s\,(2r^2/R^2 - 1) \qquad (2)$$

The observed velocity V_0 of a particle is equal to $V_e + V_w$, where V_e is the electrophoretic velocity, hence:

$$V_0 = V_e - V_s + V_s\,(2r^2/R^2) \qquad (3)$$

so that the velocity of a droplet is a linear function of the square of its distance from the axis of the tube.

Now $V_w = 0$ when $2r^2 = R^2$ or $r = R/\sqrt{2}$ (stationary level condition). Hence $r = 0{\cdot}707R$ from the axis or $0{\cdot}293R$ from the wall of the tube.

To ensure that all readings are taken at the stationary level, either that level alone is illuminated or, as in the present equipment, the microscope is set so that only the particles at the stationary level are in focus.

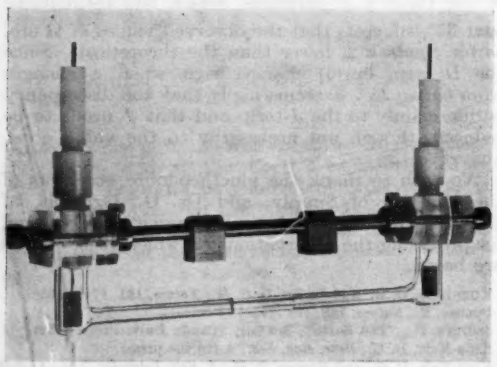

Fig. 1. Microelectrophoresis cell in bar holder with platinum electrodes in position

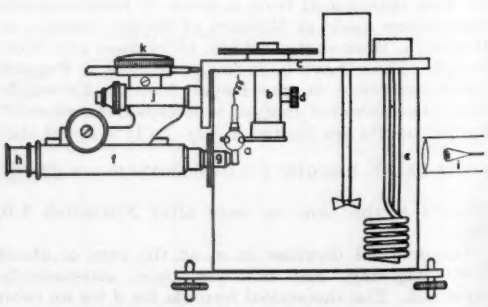

Fig. 2. Microelectrophoresis apparatus. *a*, tube holder; *b*, calibrated vertical traverse; *c*, crossbar; *d*, locking screw; *e*, thermostat; *f*, microscope tube; *g*, objective; *h*, micrometer eyepiece; *i*, light source; *j*, microscope fine adjustment; *k*, dial test indicator

Henry's correction[10] applied to the chamber of 'Veridia' glass (refractive index 1·47) amounts to 0·12 R when focusing is performed in air and is reduced to 0·01 R by immersing the objective in water.

Because of its rotational symmetry the mathematical expression of the velocity distribution for a cylindrical tube is more exact than that for the rectangular chamber where no theoretical correction for perturbation in the electro-osmotic flow produced by the side walls is made. In addition, symmetrical exchange of heat across the wall of the tube will tend to minimize convection currents.

The apparatus is illustrated in Figs. 1 and 2.

The Microelectrophoresis Tube

The tube consists of a cylindrical precision capillary of 'Veridia' glass; internal diameter uniform to ± 0·01 mm. The exact internal diameter chosen depends on the working distance of the objective; a diameter from 2·0 to 3·0 mm. is suitable for use with most × 40 objectives. At less than 1·5 mm. diameter the change in mobility of particles with depth in the tube is very large in the stationary level region, leading to large errors in mobility determinations for only small setting errors. Thermal convection effects are readily produced in tubes greater than 3·0 mm. in diameter because of the greater current flowing per unit field strength. A capillary tube of about 150 mm. was chosen since this length is convenient for levelling the cell accurately. There is no risk of contamination by electrode products since a current flowing for five times as long as is required for a mobility determination leads to contamination of approximately only the first 2·0 mm. of each end of the capillary.

'Quickfit and Quartz' *B*10 'Pyrex' sockets were fused to each end of the capillary tube and the viewing region formed by grinding and polishing a flat on the capillary wall. The capacity of the apparatus is approximately 5·0 ml. For the examination of very small quantities a similar tube fitted with *B*7 joints with a capacity less than 2·0 ml. is used. Electrodes of grey platinum[13] are inserted into the *B*10 'Pyrex' sockets. They consist of platinum foil of dimensions 12·5 × 20·0 mm., so that the current density is sufficiently low to obviate serious risk of polarization. The foil is formed into a cylinder and welded on to platinum wire which is inserted into a reservoir of mercury through a *B*10 soda glass or polyethylene plug. Reversible silver/silver chloride or copper/copper sulphate electrode systems were not used because of their greater complexity and the risk of contamination of the suspension with copper or silver salts. For currents less than 2·0 m.amp. reliable results can be obtained from these irreversible electrodes.

The Tube Holder

The tube is fixed horizontally in a holder (in Fig. 2,*a*) attached to a calibrated vertical traverse (*b*) (reading to 0·01 mm.) mounted on a crossbar (*c*) on the water-bath. A locking device (*d*) is incorporated in the adjustable traverse. Both tube and holder are immersed in a glass or 'Perspex' water-bath, the temperature controlled to ± 0·1° with an immersion heater (500 kW.) and a toluene regulator and 'Sunvic' switch or any other thermostatic device (*e*). The tube is set so that its capillary lies parallel to the surfece of the water in the bath. Cleaning is effected *in situ* using a polyethylene cannula and a suction pump. When systems of low ionic strength are used, chromic acid cannot be employed with safety as a cleaning agent unless time is allowed to leach the adsorbed chromate ions from the glass wall of the cell. A molar solution of sodium chloride is used for rinsing out between determinations at low ionic strength ($< 0·01$).

The Microscope

The microscope tube, 160 mm. long (Fig. 2,*f*), attached horizontally to the crossbar (*c*), is fitted with a × 40 ⅓-in. waterproofed objective numerical aperture 0·7 (*g*), and a × 10 micrometer eyepiece (*h*) incorporating a chessboard graticule (5 × 5 squares of 0·25 mm. side). The optical system gives a magnification of 400 diameters and is calibrated against a 1·0 mm. standard micrometer scale divided to 0·01 mm. The depth of focus is about 0·005 mm.

A light source (*i*) is conveniently mounted behind the water-bath at right angles to the tube and co-axial with the objective. The objective is inserted into the bath through a water-tight rubber gasket and can be focused on the optical flat of the tube or on the stationary level using the horizontal fine adjustment of the microscope (*j*).

An adjustable zero dial test indicator (*k*) reading to 0·002 mm. is mechanically linked to the fine adjustment of the microscope (*j*) and bears on the crossbar (*c*) to give rapid and accurate reading of the objective setting.

The rate of movement of the particles is found by timing them (stop-watch reading 0·1 sec.) over a fixed number of graticule divisions selected so as to give a reading of about 10 sec. at an applied potential gradient of 3–5 V. cm.$^{-1}$. The direction of the current is reversed half-way through each reading to minimize polarization. A mobility value is normally based on determinations on ten individual particles and is expressed in μ sec.$^{-1}$ V.$^{-1}$ cm.$^{-1}$.

Circuit

This is shown in Fig. 3. A 150 V. radio battery supplies the potential through an on/off switch to the 25 k linear potentiometer (*P*1) which acts as a potential divider. (*V*) is a Braum 0–100 μamp. centre–zero micro-ammeter with a top grade 1·0 megohm resistor (*R*) and a 100 k linear carbon potentiometer (*P*2) in series to give a 0–200 V. direct-reading high-resistance voltmeter. (*A*) is a Pye 'Scalamp' (7901/5) shunted galvanometer, sensitivity 15·6 mm./μamp. on direct reading position. The two-way double pole switch (*D*) is wired for reversing the polarity of the electrophoresis cell, but not that of the meters in the circuit.

The potential gradient was obtained directly using the applied potential and the distance between the electrodes and indirectly using the relationship field strength $F = I/KA$, where A is the cross-sectional area, I is the current passing through the tube, and K the specific conductivity of the dispersion. The two methods were found to be in excellent agreement for the systems tested.

Experimental

Since the electrophoretic mobility for washed human and sheep erythrocytes (0·1 per cent v/v suspension) in 0·145 M sodium chloride with added 0·145 M hydrochloric acid or sodium hydroxide is

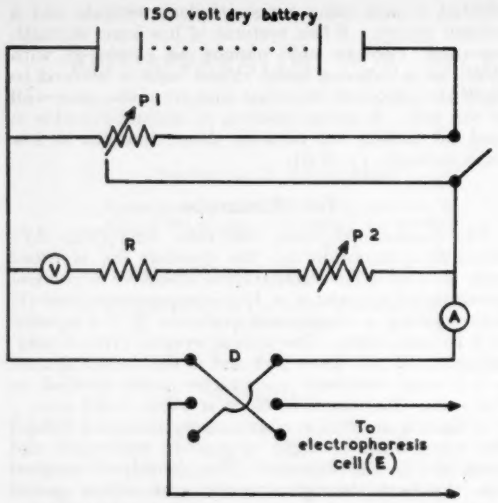

Fig. 3. Circuit diagram. *P*1, 25 *k* linear potentiometer; *P*2, 100 *k* linear potentiometer; *R*, 1 megohm resistor; *A*, galvanometer; *V*, centre-zero high-resistance voltmeter; *D*, reversing switch

constant over the *p*H range 5·0–9·0 [14], it is possible to locate the stationary level experimentally as the point of intersection of a series of *p*H depth-mobility curves (see Fig. 4). For the cylindrical tube it is also found that the relationship between velocity of the particle and the square of the displacement from the axis is linear, as predicted by equation 3 (see Fig. 5).

The stationary level is thus found by calculation and experiment and the mobility of human red cells in phosphate buffer *p*H 7·4 (2·0 ml. *M*/15 KH_2PO_4 + 8·0 ml. *M*/15 $Na_2HPO_4.12H_2O$) was 1·28 ± 0·03 μ sec.$^{-1}$ V.$^{-1}$ cm.$^{-1}$ and in 0·145 *M* sodium chloride solution it was 1·07 ± 0·02 μsec.$^{-1}$ V.$^{-1}$ cm.$^{-1}$. Table 1 shows a selection of the most reliable results obtained by other authors for the phosphate and chloride systems. Other observations in the literature do not deviate by more than about 5 per cent from those quoted, if those which can be traced to some error in technique are excluded. The difference between the chloride and phosphate values is attributed to greater anion absorption in the partially divalent phosphate case. The results

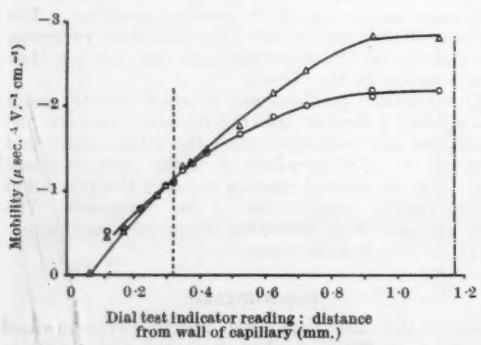

Fig. 4. *p*H-depth–mobility curves for human red blood cells. △, *p*H 9·0; ○, *p*H 6·5; ——————, stationary level; —·—·—, axis of tube

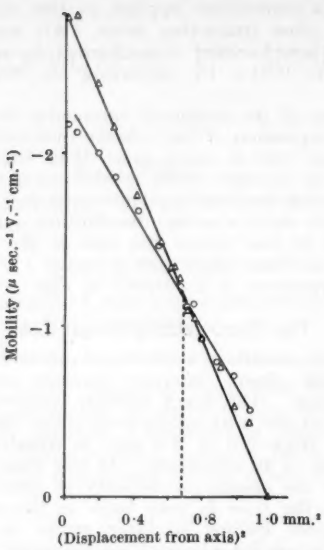

Fig. 5. Mobility–(displacement from axis)^2 relationships for human red blood cells: △, *p*H 9·0; ○, *p*H 6·5; – – –, stationary level

under the above conditions are extremely reproducible and are independent of race, sex, age or blood group.

Table 1. Electrophoretic Mobility of Human Red Cells at *p*H 7·4 and 25° C.

Ref.	Electrophoretic apparatus	Suspending medium	Ionic strength	Mobility μ sec.$^{-1}$ V.$^{-1}$ cm.$^{-1}$
2	Rectangular (horizontal position)	Phosphate	0·172	1·30
3	Rectangular (lateral position)	Phosphate	0·172	1·27
4	Rectangular	Phosphate	0·172	1·31 ± 0·02
15	Cylindrical	Phosphate	0·172	1·32 ± 0·05
16	Rectangular	Phosphate	0·172	1·28
16	Rectangular	Chloride	0·145	1·07 ± 0·02
17	Rectangular	Chloride	0·145	1·10

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PERCHLORATE, AN IMPORTANT CONSTITUENT OF SEA WATER

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PERCHLORATES, together with iodates and chromates, are known to occur in the Chilean nitre-beds[1-4]. Goldschmidt[3] suggested that these "final products of oxidative weathering" might also occur in other nitre deposits and, perhaps, in the evaporite environment in general. It now appears that both iodates and perchlorates are of widespread occurrence in the natural environment. This article is confined to the consideration of the perchlorates.

The reason why the presence of these oxyhalides has hitherto escaped attention must be ascribed in part to the inadequacy of the analytical methods[5-9]. We have tested most of the reagents described in the literature and the reproducibility of their reactions. Many of the tests for perchlorate suffer from grave disadvantages and have not been sufficiently investigated. There are reagents, like copper pyridine[10], that may yield positive results with pure perchlorate solutions but are of no use when the sample contains high concentrations of other salts. Some reagents seem to be of variable composition. This is particularly the case for reagents which are oxidized by perchlorate, such as α-naphthol, resin of guaiac (guaiacol is always inactive), *p*-phenylene-diamine and hæmatoxylin[11]. It is difficult to obtain two samples of these reagents that react in a similar fashion with perchlorate. Perchlorates are reported to react with amines. We found that no insoluble compounds were formed either with hydroxylamine or with hydrazine. It is possible that perchlorates will enter into combination with the amines occurring in the estuarine environment[12]. About most of the other reagents we will remain silent, as it would take too long to discuss their respective disadvantages[5,6].

After much trial and error we were left with three adequate presumptive tests and with two quantitative methods.

(*a*) *The 'char-reaction'*. In drying paper chromatograms it was discovered that perchlorates, in an acid environment, lower the char-temperature of the filter paper. With Whatman paper No. 544 the following results were obtained:

	Char temperature	Time
No perchlorate	230° C.	Instantaneous
Perchlorate	170° C.	Instantaneous
Perchlorate	160° C.	15 sec.
Perchlorate	105° C.	12 min.
Perchlorate	85° C.	35 min.

Among other substances tested, only chlorates and copper ions behaved in a similar way. As chlorates were never found in the natural environment, copper alone might interfere in special circumstances. For a spot-test, the filter paper is pre-treated with a saturated solution of oxalic acid, and dried. A drop of the sample solution is spotted on to the paper, and if, after heating, a jet-black ring is formed the presence of perchlorate is indicated. In the presence of high concentrations of chloride ion the central area of the spot assumes a reddish-brown colour, which is readily distinguished from the black ring given by perchlorates. The spot test may be applied at concentrations of perchlorate greater than about 100 p.p.m.

Circular chromatograms on 10-cm. paper disks (Whatman No. 41), using alcoholic ammonia according to Lederer and Cohen[9], show, on heating, a black, well-defined ring in the presence of perchlorate at R_F 0·83–0·87, the position agreeing with that obtained by other reagents. Using 0·1-ml. drops, the sensitivity may increase to less than 100 p.p.m.

(*b*) *Formation of strychnine perchlorate*[5,6]. Strychnine sulphate crystals, when placed in a dilute solution of perchlorate, slowly become covered with a felt of hair-like prismatic crystals of the perchlorate. According to the literature, this reaction is specific. If the perchlorate concentration is too high, the strychnine sulphate crystals may become immediately and completely covered with the precipitate, thus blocking further reaction, and the typical appearance is not developed.

(*c*) *Methylene blue*[5,6]. Pure methylene blue chloride, free of zinc, yields with perchlorates a purple, crystalline precipitate. Persulphates (absent in the natural environment) and certain salts of heavy metals may interfere. Although not as specific as the strychnine reaction, it is extremely sensitive and requires only small amounts of perchlorate. The methylene blue reaction has been used in the quantitative determination of perchlorate. There are many difficulties. The results reported here were obtained by using: (1) high dilutions (40 p.p.m.) of freshly prepared pure methylene blue, allowing 20–30 min. for the reaction to become complete, and filtering off the precipitate on a sintered glass filter funnel; (2) equal volumes of 25 p.p.m. methylene blue and sample, allowing 30 min. for the reaction to be completed, after which the solution is centrifuged for 10 min. at 3,000 r.p.m. The methylene blue in the filtrate is determined colorimetrically with a Spekker absorptiometer, using a red filter. Less than 100 p.p.m. perchlorate may be determined in this way, which is very satisfactory when compared with other methods[6]. Further refinement may be obtained by shaking out the molybdenum perchlorate with *iso*propyl–methyl ketone. 1 p.p.m. potassium perchlorate may be detected in this way, using a 'Unicam' spectrophotometer. In the presence of chloride the distribution of the dye between the aqueous and the ketone layer is very slow; it takes several days before equilibrium is established. For this reason, the method has not been further investigated.

(*d*) Fusion with sodium carbonate will convert perchlorate to chloride, which can then be titrated with silver nitrate[13]. This method is suitable only for samples initially low in chloride ion. The removal of the initial chloride ion by precipitation with silver nitrate appears unsatisfactory when small amounts of perchlorate are involved.

(*e*) *Other methods*. No polarographic wave is obtained with perchlorate[14]. No X-ray patterns could be obtained with natural materials or with their concentrates. Goldschmidt[3] points out that in the natural evaporite salts the perchlorate ion probably enters the crystal lattice of gypsum and that this is the reason why no specific perchlorate mineral has been found, although it may be present in amounts

of more than 6 per cent in the nitre[13]. Apart from this 'intralattice' effect in gypsum, perchlorates have a tendency to form 'interlattice' combinations with other salts, notably with sodium chloride. As no sufficiently insoluble inorganic perchlorates are known (potassium perchlorate is still fairly soluble), it seems difficult, if not impossible, to enrich a salt sufficiently to obtain an X-ray pattern of a perchlorate. Thus far we have not discovered potassium perchlorate crystals in natural waters, salts or concentrates. However, the strychnine compound could be prepared from a concentrate of a bittern from Dry Creek Salt Works (Imperial Chemical Industries, Ltd.), South Australia, supplied by Mr. C. W. Bonython. By comparing the X-ray pattern of this compound with strychnine perchlorate their identity was established by Mr. W. Roberts, of the Bureau of Mineral Resources. Inasmuch as the chemical tests on this brine concentrate are in harmony with the X-ray findings, we may now assume that substances yielding the presumptive tests described above are indeed perchlorates.

Occurrence of Perchlorates in Nature

The methods discussed above allowed us to establish the order of magnitude of the perchlorate content of various materials (Table 1). The quantities are often considerable—up to 5,600 p.p.m. in Lake Eyre brine, for example.

This is still less than one-third of the saturation value of potassium perchlorate at 20° C. Much analytical work has to be done before more reliable quantitative results are obtained.

Earlier in our work we obtained comparative results on chromatograms with *p*-phenylene diamine, hæmatoxylin and resin of guaiac for various sea-water samples.

After dissolving the blue oxidation product of guaiac resin (at R_F 0·83–0·87) in acetone, the red extinction was measured with a Lange photometer.

The results (Table 2) would suggest that the perchlorate content of sea-water tends to be highest near the coast and that it may decrease far from land and at great depths.

Table 1. Concentration of Perchlorate Ion in parts per million in various Materials

Not detected	
Tapwater (Sydney and Canberra)	
Rainwater (several samples)*	
Lake Dobson (fresh). Tasmania	
Wombat Moor (fresh). Tasmania	
Geothermal waters. Rotarua district, N.Z.	
Jenolan River, N.S.W.	
Cotter River, A.C.T.	
Murrumbidgee River, N.S.W.	
Upper Hawkesbury River, N.S.W.	
10–100 p.p.m.	
Sea-water far from land (80 miles E. of Lord Howe Island)	
Sea-water, 500 m. and 1,000 m. deep (same locality)	
Lagoon, White Island, Bay of Plenty, N.Z.	
Chlorinated distilled water	
Chlorinated tap water	
Many chemicals, analytical grade	
100–1,000 p.p.m.	
Estuarine and inshore waters (N.S.W. coast, Victoria coast, New Caledonia coast)	
Coral reefs, Gouara, Oundjoh, Isle Charron (New Caledonia)	
Mirabilite from Mawson Base, Antarctica (supplied by Mr. P. W. Crohn, A.N.A.R.E.)	
Frome River (South Australia) flowing from a deep bore, pool with algae	
Certain chemicals, analytical grade	
1,000–10,000 p.p.m.	
Salt from Dry Creek, South Australia salt works.	
Salt from Lake Eyre, 3–6 in. deep and 11 ft. 3 in. deep. Brines from Dry Creek and from Lake Eyre. Salt from Pink Lakes (Victoria), stored since 1936	
Saltpetre, potassium bromate, commercial borax, commercial salt (solar)	

* A faint trace in one sample

Table 2

Locality	Perchlorate (p.p.m.)
Gunnamatta Bay, Cronulla, N.S.W. Surface, in kelp beds	200–300
Gibbon, Cronulla, N.S.W. mouth off Port Hacking River, 100 m. deep	300
Ulladulla, N.S.W. Inshore, surface	150
Open sea, 80 miles E. of Lord Howe Island (from a cruise of the F.R.V. *Derwent Hunter*)	
Surface	100
100 m. deep	100
500 m. deep	30
1,000 m. deep	60

The conversion of the comparative figures to absolute figures is uncertain

Perchlorate Cycle

There is no reasonable doubt that iodide, bromide and chloride enter into a cycle forming, at one stage or another, higher oxidation levels. It has been unfortunate that in the literature the formation of the known oxyhalides seems to have been linked almost exclusively with the nitrogen cycle[2,3]. While nitrogen compounds may play a part in the formation of oxyhalides, there are many other ways in which these compounds may be formed. Although speculation on this matter seems premature, we should like to suggest one possibility which is in harmony with the data at our disposal.

The almost invariable occurrence of iodate (in particulate form) together with the perchlorate in both sea-water and salt suggests that the cycles of these halides are interconnected. We believe this iodate to be of biological origin, but as yet have not sufficient evidence to say this with certainty. Iodate formed directly from iodide or from elemental iodine may react with chloride to yield chlorine[4], some of which may then form perchlorate. We have found that, in the latter connexion, chlorinated distilled water contains perchlorate.

In sea-water the perchlorate is chiefly dissolved, but while none is held by a millipore filter (diameter 0·5μ), some perchlorate is retained by a Seitz asbestos filter. It may have been adsorbed by the asbestos, however. All the iodate is held by the millipore filter.

We are better informed on the breakdown of the perchlorate, at least in fresh-water. A butyric acid fermentation on malt reduced more than half a perchlorate solution (1,000 p.p.m.) in two weeks at 20° C. In the same time, a sulphate reduction brought a similar solution to 85 per cent of its original strength. Even in surface plankton swarms there may be foci of low potential in which this reduction could take place, as Mr. E. J. F. Wood (Commonwealth Scientific and Industrial Research Organization) has isolated both sulphate reducers and purple bacteria from surface phytoplankton (private communication).

Importance of the Halogen Cycle

(*a*) *Physiological.* Chili saltpetre, containing more than 0·5 per cent perchlorate, has long been known to be toxic to plants[13]. Experiments on the influence of perchlorates on growth and on germination are in progress.

(*b*) *Hydrology and oceanography*[15]. The potential variability of the perchlorate in sea-water may influence the chloride content. It has been observed repeatedly (C. Walker, Commonwealth Scientific and Industrial Research Organization, private communication) that the chlorinity of sea-water increased in the second decimal of the pro-mille chlorinity figure after storage. The order of magnitude of per-

chlorate in sea-water is such that reduction of the perchlorate may even influence the first decimal place.

It may become necessary to prepare sterile standard sea-water from perchlorate-free salts. Stored sea-water should remain in contact with the air so as to prevent the creation of low oxidation potentials and the possible reduction of the perchlorate.

Permanganates and persulphates being absent in Nature, the perchlorate represents the highest oxidation-level of the inorganic environment. While the ion is comparatively inert, it may transfer its oxygen to various phenolic compounds. It may combine with amines present in the natural environment[12], and there is reason to assume that, given sufficient time, it might transfer this oxygen to inorganic substances as well.

Any study on oxidative weathering in saline environments has to take this possibility into account.

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FAILURE OF SORBITOL TO REPLACE INTRINSIC FACTOR IN THE GASTRECTOMIZED RAT

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EVIDENCE has recently been presented that 'Liptril', a pharmaceutical preparation free of gastric intrinsic factor, may enhance the absorption of vitamin B_{12} from the gastrointestinal tract of man[1] and of the rat[2]. 'Liptril' contains vitamin B_{12} as well as various other vitamins, minerals, preservatives, flavours, and colouring material. However, Greenberg[2] and Chow[1] and their associates are in agreement that the substance responsible for the increased absorption of vitamin B_{12} is D-sorbitol, present in a concentration of 28 per cent. A similar effect was also noted in rats with certain other sugars and sugar alcohols[2].

Greenberg and his associates[2] suggest that the carbohydrate portion of the intrinsic factor may be important in vitamin B_{12} absorption. In the gastrectomized rat measurement of the effect of rat intrinsic factor may conveniently be made[3] with a dose of 0·015 μgm. of vitamin B_{12} labelled with cobalt-60. On the other hand, in the studies of Greenberg and his associates[2] with D-sorbitol on normal rats a much larger dose of the labelled vitamin (1·36 or 1·4 μgm.) was employed. Likewise, Chow and his associates[1] used 50 μgm. instead of the usual 0·5–2·0 μgm. in the Schilling urinary excretion test employed by them in demonstrating the activity of D-sorbitol in man. It therefore appeared to be of interest to learn whether D-sorbitol is effective in the rat, with or without intrinsic factor, in increasing the absorption of a small dose of vitamin B_{12}. Appropriate studies were accordingly undertaken on intact rats, as well as on rats previously subjected to total gastrectomy.

The techniques of total gastrectomy, administration of test materials, and collection and assay of faeces for radioactivity were as described by Nieweg and his associates[3]. Male rats of the Sprague-Dawley strain, weighing 200–250 gm., were subjected to total gastrectomy and allowed to recover for several days from the operation. Normal male rats of similar weight and of the same strain were employed in other experiments. The test solutions were administered by stomach tube to fasting animals, stools were collected for 96 hours thereafter, and the amount of radioactivity excreted was determined and expressed as a percentage of that administered. The test solution consisted of 1 or 2 ml. of a modified, partial aqueous formulation of 'Liptril' prepared without vitamin B_{12} and containing 28 per cent by weight of D-sorbitol, together with 0·5 ml. of water containing 0·015 μgm. of labelled vitamin B_{12} with a specific activity of 565 μc. per μgm. The control solution consisted of 2 ml. of isotonic saline and 0·5 ml. of the same solution of the vitamin.

It was soon noted that diarrhœa was frequent in the normal animals receiving the 2 ml. dose of modified 'Liptril', free of vitamin B_{12} (hereafter called modified 'Liptril') and was invariable in the gastrectomized animals receiving this dose. Consequently, results obtained with animals with diarrhœa are excluded, and in the gastrectomized animals only those obtained with 1 ml. of modified 'Liptril' are reported. No attempt was made to separate the animals which had received modified 'Liptril' from those given isotonic saline as a control, and so a number of the animals received both preparations separated by an interval of at least two weeks.

As shown in Table 1, the normal rats excreted about 50 per cent, the gastrectomized rats essentially all, of the radioactivity of the dose of labelled vitamin B_{12} administered. No certain differences were noted between the average fæcal excretion of radioactivity by those animals receiving modified 'Liptril' and by those receiving saline. This lack of effect applied to both the intact rats and the gastrectomized animals.

In the technique employed here, all or nearly all the radioactivity of a dose of 0·015 μgm. of vitamin B_{12} labelled with cobalt-60 is excreted by the gastrectomized rat[3]. However, the simultaneous administration of 3 ml. of rat gastric juice or of one-half of a

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Table 1. Fæcal Radioactivity as Percentage of Oral Dose of Radioactive Vitamin B_{12}

Animal type	No. of experiments	Vehicle for Co^{58}–B_{12} 0·015 μgm.	Fæcal radioactivity (per cent ± 1 S.D.)
Intact rat	12	D-Sorbitol elixir*	43·7 ± 12·0
,, ,,	12	Saline	44·0 ± 6·7
Gastrectomized rat	5	Modified 'Liptril'	99·0 ± 10·4
,, ,,	10	Saline	93·6 ± 8·3

* Modified, partial, aqueous formulation of 'Liptril' prepared without vitamin B_{12} and containing 28 per cent of D-sorbitol.

homogenized rat stomach will reduce the fæcal excretion of radioactivity to approximately that of the normal rat, about 50 per cent of the dose administered. The absence of any such effect with modified 'Liptril' in these gastrectomized rats suggests that, even at the maximum dose tolerated without diarrhœa by the animals, it did not take the place of rat intrinsic factor; nor did twice this amount of modified 'Liptril' enhance the absorption of the small dose of vitamin B_{12} by the intact animal. On the other hand, 'Liptril' free of vitamin B_{12} or D-sorbitol apparently augments absorption of vitamin B_{12} by the intact rat when approximately 100 times as much vitamin is administered[2].

In pernicious anæmia Ross and his associates, and recently Doscherholmen and Hagen[4], have demonstrated that two mechanisms seem to exist for the absorption of vitamin B_{12}. Rapid absorption follows a rather large dose of the vitamin (50–3,000 μgm.) and is independent of or even inhibited by intrinsic factor. On the other hand, a slower, apparently physiological mechanism, dependent upon the presence of intrinsic factor, is required for the absorption of small amounts of vitamin B_{12} (0·5–1·0 μgm.). The substance in 'Liptril' promoting the absorption of large doses of vitamin B_{12} in the rat[2] and in man[1] has been shown to be D-sorbitol. Consequently, those reports, as well as the present experiments, suggest that D-sorbitol affects the rapid mechanism for the absorption of vitamin B_{12} in man described[4] as independent of intrinsic factor activity. Further evidence for this is found in the ability of D-sorbitol and of certain other carbohydrates to enhance iron absorption in both the rat and man[5] as well as calcium absorption in the rat[6]. Other observations in patients underline the importance of the size of the dose of vitamin B_{12} in determining the ability of D-sorbitol to enhance its assimilation. Thus, Chow and his associates failed to observe such an effect in pernicious anæmia when 2 μgm. of radioactive vitamin B_{12} were given during remission, whereas Helen E. Duell[7] has observed enhanced hæmatopoietic activity with D-sorbitol in a few such patients in relapse when the dose of vitamin B_{12} was 15 μgm.

The carbohydrates reported to be active in promoting the absorption of vitamin B_{12}, calcium, and iron in the rat seem to be those classically considered to be absorbed by passive diffusion from the intestine, whereas carbohydrates considered to be absorbed by phosphorylation, such as gluctose and fructose, are reported to be inactive[2]. This perhaps further suggests that the action of D-sorbitol does not involve a specific transport mechanism.

We are indebted to Mrs. Felicita Fradera for technical assistance, to Dr. S. M. Greenberg, of Smith, Kline and French Laboratories, for D-sorbitol, and to the Merck, Sharp and Dohme Research Laboratories for the labelled vitamin B_{12}.

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DEGRADATION OF BLOOD GROUP-SPECIFIC MUCOPOLYSACCHARIDES BY FICIN AND PAPAIN

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Mucopolysaccharides composed of carbohydrate and amino-acids firmly bound together are responsible in man for immunological specificity within the ABO and Lewis blood group systems[1]. The specific substances are usually isolated by extraction of freeze-dried secretions or tissue digests with cold 90 per cent phenol solution, the serologically active mucopolysaccharide remaining largely insoluble in this solvent whereas the protein impurities pass into solution[2]. By the use of this simple procedure, followed by fractionation of the specific materials from aqueous solution by organic solvents and/or inorganic salts, it has been possible to isolate and study the properties of the human blood group substances[3]. In spite of the different serological characters of the A, B, H and Le^a substances, they each possess the same qualitative composition and contain the same four sugars—L-fucose, D-galactose, D-glucosamine and D-galactosamine, and eleven amino-acids, none of which is aromatic or contains sulphur.

Proteolytic enzymes have also been used to remove adventitious protein during the isolation and purification of the group substances from materials of human and animal origin, and it has generally been accepted that the specific mucopolysaccharides are not changed by such treatment[4]. A few early observations[5], however, suggested that in certain instances a destruction or modification of the serological character occurs, but where this has been reported the modified substances were not recovered and the physico-chemical changes induced were not determined.

A study of the literature reveals that, almost without exception, the preparation of highly purified specific mucopolysaccharides has involved at some stage treatment with 90 per cent phenol. An attempt was therefore made to isolate the specific substances by a method[6] which did not involve the use of

phenol, and to achieve this the native secretions or tissues were incubated with activated ficin at about *p*H 7. It was hoped by this means to remove the accompanying unspecific protein and ultimately to recover unchanged the specific substances. Mucopolysaccharides recovered after treatment with ficin, however, gave solutions of much-reduced relative viscosity, as compared with the materials isolated by means of simple phenol extraction. The ficin-treated group substances were readily soluble in phenol and even soluble to some extent in liquid 95 per cent w/w phenol, whereas specific substances recovered by extraction of the original materials with 90 per cent phenol were largely insoluble in this solvent. The behaviour of relatively homogeneous specific substances on ultracentrifugation and electrophoresis at *p*H 4 and 8 suggested that after ficin treatment they became polydisperse and contained more than a single component. These marked differences in properties indicated that an alteration in the structure of the specific mucopolysaccharides had occurred, but no definite evidence was obtained at that time that the serological properties of the materials were modified.

A closer examination of the substances treated with ficin with those recovered from the same source by simple phenol extraction revealed that the specific substances after treatment with ficin have a lower capacity to neutralize the corresponding antibody in hæmagglutination tests. This reduced activity is not always obvious when cyst fluids or other secretions are given a single short treatment with ficin at *p*H 7, especially when they contain, as they frequently do, a large excess of unspecific protein.

These additional findings prompted a more detailed study of the action of ficin on preparations of specific substances which had been extensively purified by several successive extractions with cold 90 per cent phenol until their analytical composition remained unchanged. To remove possible minor contaminants which might remain, the substances extracted with phenol were fractionated further with ethanol and a fraction, which amounted to about 95 per cent of the whole, separating between 43 and 45 per cent ethanol concentration, was collected. Purified ficin was obtained from dried commercial fig latex by dialysis and fractionation with ammonium sulphate[7]. The material was electrophoretically homogeneous and behaved as a single substance when eluted from a column of Amberlite 1*RC*–50 with 0·2*M* phosphate buffer (*p*H 5·6). Experiments were also made with crystalline ficin.

A typical hydrolysis experiment was carried out as follows: thoroughly purified *A* substance dissolved in phosphate-citrate buffer at *p*H 4 was incubated at 37° for 16 hours with about 1 per cent w/v ficin, which had been activated with 2 : 3 dimercaptopropanol (B.A.L.). Toluene was added as a preservative. The solution was then saturated with ammonium sulphate, filtered, dialysed free from salt, evaporated under reduced pressure and dried from the frozen state. The treatment with ficin was repeated and the final product was fractionated from 2 per cent w/v solution in 2 per cent w/v sodium acetate solution, by the addition of ethanol. The major part of the substance separated between 47 and 55 per cent v/v ethanol, whereas the original material under similar conditions precipitated completely between the levels 43 and 45 per cent v/v ethanol. The substance treated with ficin was found to be closely similar in composition to the material before treatment, but possessed no more than about 3–6 per cent of the activity of the original A substance, when measured in the same test against natural human anti-A agglutinin. The value for activity showed a smaller reduction when immune rabbit or human serum was used, but the final figure attained varied somewhat according to the specimen of anti-A serum used. Materials separating at higher (50–60 and 60–75 per cent v/v) concentrations of ethanol were relatively small in amount but gave similar analytical figures, and showed an even lower serological activity. Rabbit anti-A serum and pneumococcus type XIV horse anti-serum precipitated strongly when mixed with an equal volume of a 0·1 per cent solution of all these mucopolysaccharide fractions. There was no evidence that the capacity of the materials to inhibit the hæmolysis of sheep cells by anti-A cell immune rabbit serum, and complement decreased significantly. The material remaining in solution in 75 per cent ethanol possessed about 0·2 per cent of the activity of the original A substance; nevertheless, it remained closely similar to the original substance in chemical composition. Typical analytical figures obtained for the products arising from group A substance are given in Table 1. Ficin brings about the changes described over a broad *p*H range (4–7·6). Repeated treatment with ficin of the main fraction, which was precipitated from solution between 47 and 55 per cent v/v ethanol, failed to lower further the serological activity or change the analytical composition (after the fourth treatment with ficin : nitrogen, 4·3; fucose, 24·9; hexosamine, 30·9; reducing sugar, 53·3 per cent) and material soluble in 75 per cent v/v ethanol was no longer formed. Complete serological inactivation, as measured by precipitation and hæmagglutination inhibition tests, however, can be readily brought about by the action of an enzyme preparation obtained from *Trichomonas foetus*[8].

Table 1. Analytical Values for the Products of the Action of Ficin on Blood Group A Substance

	Original A substance	Fractions precipitated from ficin-treated A substance				
		Ethanol concentration (per cent)				
		47–50	50–55	55–60	60–75	above 75
Nitrogen (per cent)	5·2	4·4	4·7	4·4	4·4	3·4
Fucose (per cent)	23·4	25·5	23·8	23·4	21·5	17·4
Hexosamine* (per cent)	31·5	32·0	31·6	32·0	30·4	26·0
Reducing sugar* (per cent)	50·0	51·3	53·5	51·4	52·3	57·0
Hæmagglutination inhibition activity (μmoles)	0·5	8	8	16	64	128
Specific optical rotation $[\alpha]_D$	−50°	−30°	−43°	−42°	−46°	—
Relative viscosity at 20° in water (1 per cent solution)	4·68	2·39	1·51	1·21	1·10	—

* After the materials have been hydrolysed with 0·5 *N* hydrochloric acid for 18 hr. at 100°. Hexosamine is given as 'base'; the reducing values in terms of glucose equivalents.

Similar experiments, in which the step involving purification with ammonium sulphate was omitted and the dialysis waters were concentrated and examined by chromatography for diffusible products, showed that simple sugars and oligosaccharides were not released in demonstrable amounts from the specific substances as a result of treatment with ficin. Amino-acids and/or peptides were usually found in the diffusates, but only in small quantities. Their

appearance can be correlated with the small but consistent decrease in the nitrogen content of the material recovered after treatment with ficin, but it has not yet been finally established whether these amino-acids are derived from the specific mucopolysaccharide alone, and the loss in specific activity is due to changes induced by their release, or whether they arise from small amounts of contaminating protein or peptide in the specific substance.

Similar detailed hydrolysis experiments with ficin have been made using H substance, and its general behaviour cannot be distinguished from that described above for A substance (see Table 1). Although as yet studied in much less detail, B and Le^a substances appear to be changed by ficin in a similar manner, again with extensive but limited loss in serological activity.

The specific mucopolysaccharides have been treated with crystalline papain under similar conditions, and here again the same sequence of events has been found to occur; there was a reduction in serological activity to a value equivalent to a few per cent only of the original, and no very obvious change in chemical composition occurred. The results of some preliminary experiments indicate that papain can extend the hydrolysis brought about by ficin, which suggests that these two proteolytic enzymes can act on different peptide structures within the mucopolysaccharides.

The changes observed when ficin acts on the specific substances can be explained most simply by assuming that the materials, which are macromolecular carbohydrate/amino-acid complexes, are degraded by rupture of a limited number of peptide bonds and relatively large carbohydrate/amino-acid units are set free with no release of simple sugars or diffusible oligosaccharides. In support of this suggested mode of attack, there is other evidence which can be briefly mentioned. The extent of the changes induced in the specific mucopolysaccharide depends on the quantity of the enzyme used; definite hydrolysis can still be recognized when a substrate:ficin ratio of 1,000 : 1 is employed. The hydrolysis of the specific substances can be completely inhibited if the ficin is incubated with an equimolar quantity of *para*chloromercuribenzoate before the addition of the substrate and the activity can be restored either by adding a 6–10-fold excess of substances containing free SH groups (B.A.L., cysteine, etc.), or by dialysis of the mercury-ficin compound against 0·01 *M* cystein solution. The ficin used did not contain lysozyme, and crystalline lysozyme had no action on the specific substances as measured by the hæmagglutination inhibition test. The enzyme used did not appear to contain α- or β-galactosidases or β-N-acetylglucosaminidase.

Paramucin, mucus surface epithelium, and other water-insoluble complexes containing mucopolysaccharide can be readily brought into solution by digestion with ficin or papain at neutral *p*H, and this procedure was recently suggested as the basis of a method for obtaining the specific substances from these and other biological materials[6]. Now that it has been shown that ficin and papain bring about limited, but nevertheless important, changes in the specific materials, it is obvious that these enzymes should not be used for the isolation of the specific substances, if native mucopolysaccharide preparations are required.

If the present views concerning the structures associated with blood group specificity within the ABO and Lewis systems are correct, then it is the non-reducing end residues of certain oligosaccharide units which are mainly responsible for the serological character on which group specificity depends. As a result of thorough treatment with ficin there has been no loss from the specific substances of carbohydrate, either in the form of simple sugars or as diffusible oligosaccharide fragments, and therefore presumably the specifically reactive structures are still intact in spite of the considerable fall in inhibition titre observed. It is not surprising, therefore, to find that the major part of the materials formed as a result of the limited proteolysis react strongly in specific anti-A precipitation tests.

It is clear that the chemical changes induced in the specific substances by ficin or papain are very different from those brought about by extracts of *Trichomonas foetus*[8], many bacterial culture filtrates[9], and hepato-pancreas preparations from certain snail species[10], etc., where crude or purified enzyme systems bring about an extensive release of small diffusible sugar units together with complete destruction of the characteristic serological specificity. Treatment of the specific substances with ficin and papain, on the other hand, appears to hydrolyse only a few of the potentially susceptible peptide bonds and to give rise to a limited number of smaller fragments resistant to ficin, closely similar in composition to that of the original substance, and in this manner induce changes not brought about by any method previously described. The use of these proteolytic enzymes of plant origin and possibly of other similar ones from animal tissues opens up new possibilities for an experimental approach to the isolation of relatively small structural entities containing unchanged carbohydrate units joined to peptides and ultimately, perhaps, devoid of amino-acids, and thus constitutes a valuable tool for the study of mucopolysaccharide structure.

Full details of these experiments will be given elsewhere.

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LETTERS TO THE EDITORS

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Seasonal Illumination of a Circumpolar Earth Satellite at its Extreme-Latitude Orbit Point

Optical tracking of Earth satellites is the only means of determining their motion with such precision as to render measurements of air density significant. This, however, can only be done when the satellite is sunlit, considering that a flashing beacon (which has been suggested) would relatively quickly run out of power. I shall discuss the insolation of Earth satellites revolving on orbits similar to those of the Russian *Sputnik*.

Table 1

P		K		$2(\pi - K)$	
Deg.	min.	Deg.	min.	Deg.	min.
084	50.0	000	00.0	360	00.0
090	00.0	036	39.5	286	40.0
100	00.0	064	07.5	231	45.0
110	00.0	085	48.7	188	22.6
113	26.5	093	10.0	173	40.0
120	00.0	108	10.7	143	38.6
125	00.0	121	31.2	116	57.6
134	49.0	360	00.0	000	00.0

Arcs $ab = 2K$ and $abc = 2(\pi - K)$ are functions of the polar distance of the Sun. Table 1 gives the values of these arcs corresponding to certain values of P.

These values are plotted in Fig. 2. The inner scale of the abscissæ gives the arc $2(\pi - K)$, and the outer scale gives the hours, H, before and after solar noon at which the summit-point would still receive the Sun's rays. When the satellite goes over that northernmost point of the orbit (for the northern hemisphere), it will catch the Sun's rays even if the rest of the orbit is in shadow. Actually, due to the refraction, the illuminated part of the orbit will be more than a point. For the northern hemisphere the summit-point of the orbit will be in constant sunlight during April 5–September 8. For the southern hemisphere these dates will be October 8–March 6.

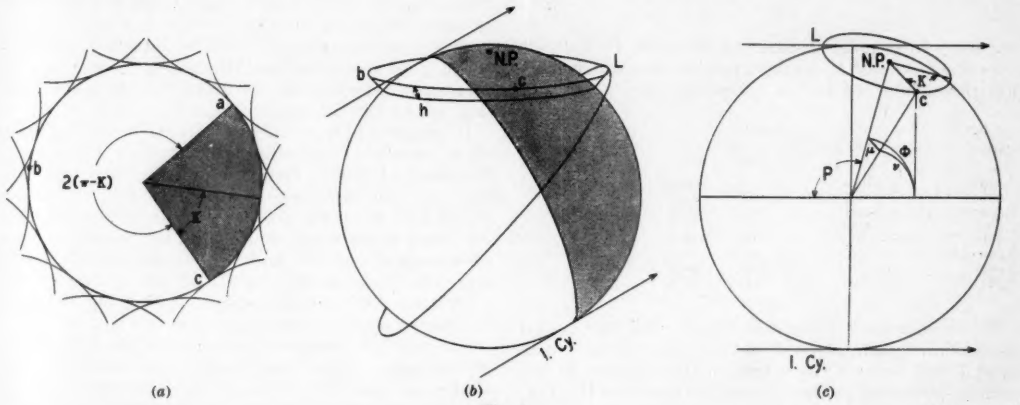

Fig. 1

The summit points of the orbit or points nearest the pole define a locus, L, which is centred on the Earth's axis, provided that the inclination of the orbit is different from zero. Verticals passing through the summit-points and their sub-points define a conical surface with its vertex at the centre of the Earth. This locus L (Fig. 1) forms the base of the cone, the intersection of which, with the illumination cylinder $I.Cy$, defines the illuminated arc abc and its complementary arc ac, which is in shadow. Let the angles of these arcs be, respectively, $2(\pi - K)$ and $2K$

Consider an orbit with inclination, $i = 65°$, and a height of passage at the summit-point, $h = 400$ km. (this is, somewhat schematized, the case of the Russian Earth satellites). Then we may derive K from the following relations and quantities (Fig. 1*c*):

$$\cos K = \frac{\cos \nu - \cos \varphi \cos \mu}{\sin \varphi \sin \mu}$$

where $\mu = (90° - i) = 25°$ and, if P is the polar distance of the Sun, $\varphi = (180° - P)$. The angle ν is given by:

$$(R + h) \sin \nu = R$$

where R is the radius of the Earth (6,350 km.) and h is 400 km. Thus:

$$\cos K = \frac{0{\cdot}33901 + \cos P \times 0{\cdot}90631}{\sin P \times 0{\cdot}42262}$$

The h-curves of Fig. 2 give the values of $2(\pi - K)$ and the hours of insolation of the summit-point as a

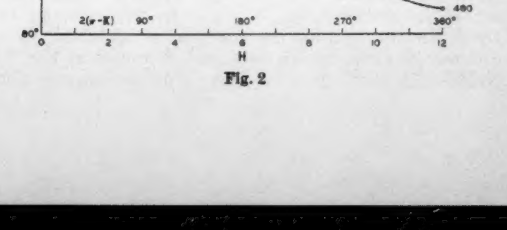

Fig. 2

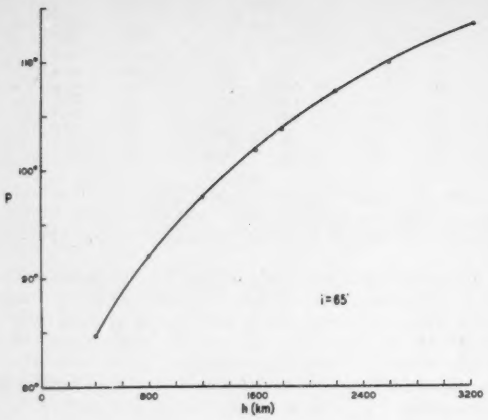

Fig. 3

function of the polar distance of the Sun, P. Table 2 gives the values of P required to give constant insolation throughout 24 hr. as a function of h.

Table 2

h (km.)	cos (90 − ν)	(90 − ν) Deg.	min.	P Deg.	min.
400	0·94074	019	49.0	084	49.0
800	0·88811	027	21.5	092	21.5
1,200	0·84106	032	45.0	097	45.0
1,600	0·79784	036	59.5	101	59.5
1,800	0·77914	038	49·0	103	49.0
2,200	0·74269	042	02.3	107	02.3
2,600	0·70950	044	48.5	109	48.5
3,247	0·66327	048	27·5	113	27.0

These values are plotted in Fig. 3. All-year-round insolation of summit-point and therefore of the entire locus L will occur if the height of the satellite at the summit-point is 3,247 km., since for this value $P = 113°$ 27′, which is the polar distance of the Sun at the winter solstice for either hemisphere. Atmospheric refraction, which will increase the illuminated arc, has not been taken into consideration.

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Interface Couples in Crystals

Joel and Wooster[1] have discussed cases in which 'volume couples' can arise in certain classes of crystals due to strains, and have mentioned that in such cases the stress tensor is not symmetrical ($\sigma_{yz} \neq \sigma_{zy}$). Actually, it is possible for asymmetry of the stress tensor to arise in any class of crystals, due to distributed couples which are acting across an interface rather than being distributed throughout a volume. These distributed interface couples have the same relation to the distributed volume couples that stresses have to distributed body forces.

Let χ_{yz} represent the component of the interface couple per unit area acting on a y-face and having its moment vector in the z-direction. For equilibrium across an interface, $\chi_{yz} = \chi_{-y-z}$. In the presence of these surface couples the moment equilibrium of a differential element can be found as noted in Fig. 1. Neglecting the effect of any volume couples for brevity, the condition for equilibrium of moments about the z-axis is:

$$\left(\sigma_{xy} - \sigma_{yx} + \frac{\partial \chi_{xz}}{\partial x} + \frac{\partial \chi_{yz}}{\partial y} + \frac{\partial \chi_{zz}}{\partial z}\right) dx\, dy\, dz = 0 \quad (1)$$

Equilibrium of the other components of moments gives similar equations. Thus the deviation from the classical theory of elasticity in this case is due to the derivatives of the interface couples. It should be noted that the moments due to stress gradients of the type $\partial\sigma_{xx}/\partial y$ are higher-order differentials.

The transformation law for the interface couples may be derived in a manner similar to that for stresses, by considering moment equilibrium of a tetrahedron. In terms of the direction cosines $l_{ij'}$ between the i- and j'-axes,

$$\chi_{i'j'} = \sum_{i=1}^{3} \sum_{j=1}^{3} \chi_{ij} l_{ii'} l_{jj'} \quad (2)$$

Thus the interface couples are tensors. It should be noted that the stresses do not become involved in the transformation law. The interface tensor is not in general symmetrical (that is, $\chi_{yz} \neq \chi_{zy}$), so that even in two dimensions the Mohr's circle transformation would not be applicable.

It seems likely that the 'strains' resulting from these interface couples are the lattice curvatures $\varkappa_{ij}$ discussed by Bilby, Bullough and Smith[2]. Relations between the interface couples and the curvatures would add 45 elastic constants. Interaction between the usual stresses and strains and the newer concepts of interface couples and curvatures, which would occur in the case of crystals of the symmetry discussed by Joel and Wooster, would bring the total number of elastic constants to $(18^2 + 18)/2$, or 171.

An order-of-magnitude estimate of the importance of interface couples in a crystal may be obtained by assuming that the atomic forces causing interface couples are of the same order as those causing ordinary stresses. The interface couple χ acting on one face of a unit cell of dimension a gives rise to a couple χa^2. The moment arm of this couple is no greater than the order of the lattice spacing a, so that $\chi a^2/a \leqslant 0$ (F). But the interatomic forces are also of the order of the normal stress, $E\varepsilon$, times the area

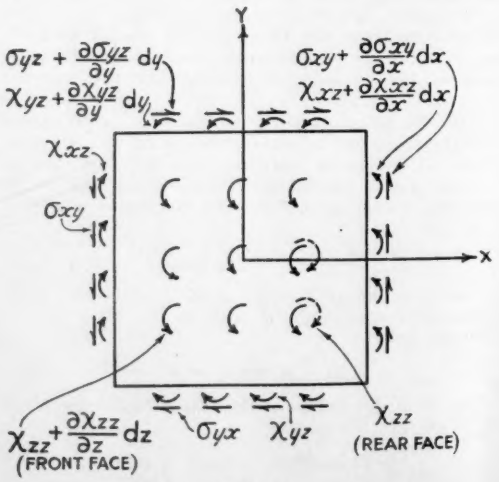

Fig. 1

of the face: $F = 0\ (E\varepsilon a^2)$. Thus the interface couples will be no greater than $\chi \leqslant 0\ (E\varepsilon a)$. For the gradients of the couples to be of the same order as the shear stresses, so that, from equation 1, $\sigma_{xy} - \sigma_{yx} = 0\ (\sigma_{xy}) = 0\ (\sigma_{xx})$, the difference of the shear stresses is of the magnitude of the stresses themselves:

$$\frac{\partial \chi}{\partial x} = 0\left(Ea\,\frac{\partial \varepsilon}{\partial x}\right) = 0\ (E\varepsilon)$$

$$\text{or}\quad \frac{\partial \varepsilon}{\partial x} = 0\left(\frac{\varepsilon}{a}\right)$$

Such high gradients of strain would be expected only around singularities such as cracks and dislocations.

Interface couples may also be of importance in laminated plastics reinforced by fibre. If alternate plies are laid with the fibres perpendicular to each other, and the whole assembly pulled at 45° to both fibre directions, there will be couples acting on the interfaces between plies wherever fibres cross over each other. The sign of these interface couples changes from one side of a ply to the other, so that there would be large gradients of the interface couples and hence $\sigma_{xy} \neq \sigma_{yx}$.

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Molecular Orbitals and Vibrational Potential Function of Nitryl Fluoride

In their work on the electronic structure of the N_2O_4 molecule, in which the N—N bond was found to be a π-only bond, Coulson and Duchesne[1] were led to consider the structure of a single NO_2 molecule. This can be represented using in particular two nitrogen lone-pair electrons, three π-electrons associated with the whole O—N—O framework and four σ-electrons. The aim of this communication is to see to what extent this picture may be used as a basis for the study of NO_2F in terms of molecular orbitals. In this molecule, contrary to what occurs in N_2O_4, N_2O_3 and N_2O_2, we should expect a normal σ-bond for NF. Such a bond should be obtained by promoting one of the lone-pair electrons to the π-orbital labelled Q by Coulson and Duchesne[1]. This gives rise to a total of four π-electrons, distributed in the P and Q molecular orbitals, whereas the remaining lone-pair σ-electron is coupled with the $2p_z$ electron of fluorine.

Let us now consider the effects which can be expected from this redistribution of electrons on the properties of the NO_2 group. As the Q orbital may be assumed to be O $\leftrightarrow$ O anti-bonding, the ONO angle has to be larger in NO_2F than in NO_2, whereas the corresponding bending force constant should be smaller. On the other hand, as the Q orbital is N $\leftrightarrow$ O non-bonding (or perhaps very slightly anti-bonding), the change in the NO bond-lengths ought to be insignificant, though the variation of the hybridization character at the nitrogen atom caused by the angle opening might give rise to a small bond shortening.

Table 1

Force constants	NO_2F	NO_2*	NF_2†
f_{NF}	4·3– 5·3		4·28–5·27
f_{NO}	11·0–11·8	10·42	
f_{ONO}	0·2– 0·5	1·099	
f_{FNO}	1·5– 1·6		

* From ref. 5.
† From values given in ref. 6.

The validity of this model must be tested, and a comparison with the vibrational potential function is valuable. (It is to be noted that the molecular dimensions reported[2] for NO_2F are affected by the fact that the value for the ONO angle had to be assumed.)

In Table 1 are given the limits between which the force constants of NO_2F (expressed in 10^5 dynes/cm.), which is of C_{2v} symmetry[3,4], are found to vary when making reasonable assumptions of the values of the coupling constants. (The accuracy for the values of the interaction terms is too low to make any suggestion here. It is, however, fairly certain that f_c(NO/NF) and γ_c(NF/ONO) are respectively positive and negative.) The results have been deduced from a general potential function, introduced earlier by Duchesne[3]. The infra-red and Raman data have been taken from the work of Dodd, Rolfe and Woodward[4]. For comparison, the corresponding constants for NO_2 and NF_2 are also given in the table.

First, it is seen that the NF stretching constants are almost the same in NO_2F and NF_2, in agreement with our model. Secondly, on going from NO_2 to NO_2F a large weakening of the bending constant (about 65 per cent) and a small strengthening of the NO bonds (about 10 per cent) are observed. This also was to be expected on the basis of the general properties of the Q orbital, the anti-bonding character of which is now estimated. The extension of these results to the discussions of molecules such as NO_2Cl and NO_2Br is premature, due to lack of experimental data. It is to be hoped that the present work, which may be compared with Duchesne's work on the electronically excited molecules[3], may indicate a fruitful way for testing and evaluating the properties of the molecular orbitals.

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An Oxidation Reaction Highly Dependent on Pressure

The rates of oxidation of metals in gases at high temperatures are frequently influenced by the pressure of the gas. The dependence on pressure, which has in some cases been theoretically explained[1], is gener-

ally small, often according to some fractional power[2]. During an investigation of the oxidation of aluminium alloys in high-temperature steam we have found a very much greater dependence on pressure.

In our experiments, the alloy "Aereal 1" (aluminium + 2·5 wt. per cent nickel), chosen for study because of its good resistance to high-temperature water[3], was exposed to dry steam at 325° C. and pressures from 550 to 1,500 lb./in.^{2} for periods up to 160 hr. Oxidation was assessed by the gain in weight of the specimen—a valid measure, since wholly adherent films were formed. Oxidation/time curves for each pressure followed a parabolic equation ($w = Kt^m$), where $m = 0{\cdot}48$. The constant K was found to be markedly dependent on pressure; assuming $K \propto p^n$, values of n from 3·9 to 4·3 were obtained, so that the parabolic rate constant was approximately proportional to the fourth power of the pressure.

Since the reaction followed a parabolic relation with time at all pressures, implying that the rates are inversely proportional to film thickness, the controlling process must be diffusion through the oxide film. It is on the latter, therefore, rather than on a surface reaction of the oxide/steam interface, that variations of steam pressure must exert their effect. No certain explanation of the effect can be offered at present, but it seems likely that, under the conditions of the experiments, the composition of the film is significantly changed by variations of steam pressure.

Studies of the equilibrium between steam and bulk aluminium oxides indicate that corundum (α-Al_2O_3) replaces böhmite (γ-AlO(OH)) as the stable phase at about 385° C.[4]. This transition temperature may be rather lower when the oxide forms as a surface film, as is true of the analogous change from bayerite ($Al(OH)_3$) to böhmite. (For the bulk substances, the transition temperature[5] is 130° C.; but, on corroding aluminium, böhmite forms the film[6] above 60° C.). The conversion of AlO(OH) to Al_2O_3 involves the elimination of water, so that at lower pressures the film composition should be nearer α-Al_2O_3, which might well be more protective than böhmite. If the experimental temperatures were near that of the transition at the pressures studied, a large dependence of composition, and thus of corrosion-rate on pressure, could occur. Preliminary results at 350° C. (and similar steam pressures) suggested not only that rates of corrosion were lower than at 325° C., indicating a more protective film, but also that the dependence on pressure was less, as might be expected further from the transition temperature.

Additional experiments, including a study of the composition of the oxide films, are proceeding and will be reported elsewhere.

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Thermal Decomposition of Hydrogen Peroxide Vapour

Knowledge of the decomposition of hydrogen peroxide vapour is of considerable importance to the developing understanding of unimolecular reactions and also of the oxidation of many gaseous fuels. McLane[1] and Giguère and Liu[2] claimed the overall homogeneous decomposition to be first-order. Satterfield and Stein[3] claimed that the decomposition occurred by a chain reaction the overall order of which was 1·5. We have carried out an extensive investigation of the decomposition and shown that it is non-chain and second-order (that is, the rate can be expressed as $k[H_2O_2]\,[M]$) up to pressures of at least 1 atmosphere of nitrogen). We have further shown that the first step in the decomposition must be the second-order reaction:

$$H_2O_2 + M \rightarrow 2OH + M \qquad (1)$$

Hoare and Walsh[4] had earlier given reasons why (1) would be expected to be second-order at ordinary pressures. The efficiency of M in bringing about reaction (1) decreases in the order $H_2O_2 > H_2O > CO_2 > N_2 > O_2 > He$.

Using our values for the efficiencies of the gases which played the part of M in the work of McLane[1], Giguère and Liu[2] and Satterfield and Stein[3], we have calculated, from the results of these authors, second-order rate constants which, after proper correction for heterogeneous effects, agree excellently one with another and with our own results. The rate constant of (1), when M is H_2O_2, is found to be $10^{15{\cdot}4} \exp(-48{,}000/RT)$ l.mole^{-1} sec.$^{-1}$.

A full account of this work will be published elsewhere.

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Deuterium Exchange in Polypeptides

Observations on the rate of deuteration of the NH groups in solutions of proteins in heavy water (see, for example, ref. 1) have shown that in some proteins there is both a fast and a slow exchange, and it has been conjectured that those NH groups which exchange slowly are hydrogen-bonded in α-helix arrangements, whereas the fast reaction is associated with solvated NH groups. This possibility can be more profitably examined in solutions of a simple polypeptide such as poly-γ-benzyl-L-glutamate, where the conditions for α-helical or random, solvated coil forms are now well-known[2,3]. Doty and Yang[3] have shown that in mixtures of dichloracetic acid and ethylene dichloride, samples of poly-γ-benzyl-L-glutamate of high molecular weight may exist either in α-helix or in random coil forms, according to the composition of the solvent.

We have followed the rate of deuterium exchange by measuring the strength of the hydrogen-bonded peptide NH band at about 3,300 cm.$^{-1}$. Since chloroform is more transparent in this region than ethylene dichloride, we have used a mixture of chloroform and O-deuterated dichloracetic acid as solvent. By measurement of the dispersion of optical rotation, we have found, as expected from the work of Doty and Yang, that with 10 per cent dichloracetic acid in chloroform (v/v) the polymer is all in the α-helix form. In pure dichloracetic acid only the random form is found[4].

If poly-γ-benzyl-L-glutamate is dissolved in O-deuterated dichloracetic acid, and chloroform is added to give a 10 per cent solution of the acid, the spectrum of the solution is found to show no NH band, even when the time required for solution and observation is reduced to less than an hour. The deuterium exchange evidently proceeds very rapidly. When, on the other hand, the polymer is first dissolved in chloroform, and 10 per cent of O-deuterated dichloracetic acid is then added, a strong NH band is seen in the spectrum; this band does not diminish appreciably in a period of days, and deuterium exchange is very slow. There is therefore no doubt that, under the conditions described with a polymer concentration of 0·5 per cent w/v, the random, solvated coil allows the deuterium exchange to proceed very much more quickly than the α-helix form. It would, however, be rash to suppose that there are not other important factors to be considered in interpreting deuterium exchange in protein solutions.

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Quantitative Separation of Nitrogenous Organic Bases using Ammonium Reineckate

During a study of the reineckates of a series of clinically important basic compounds, the mechanism of the reaction between the bases and ammonium reineckate was studied. It was found that the formation of the complexes depends on the basicity of the compounds, and that under suitable *p*H conditions certain bases can be separated quantitatively from one another. A brief discussion of the reaction mechanism on which the separation is based is presented here, and a full report on this work will be published elsewhere.

When a base (*B*) is dissolved in water the following equilibrium exists:

$$B + H_2O \rightleftharpoons BH^+ + OH^- \qquad (1)$$

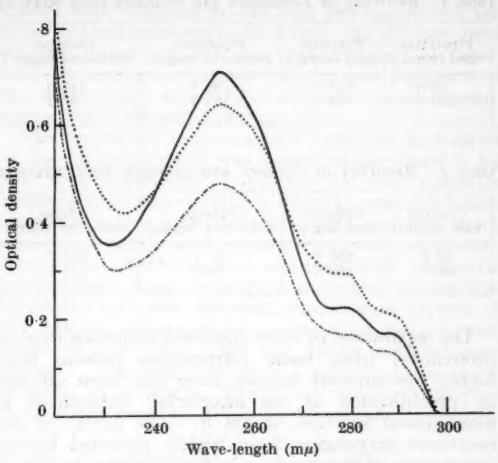

Fig. 1. Spectra of strychnine. ———, The standard strychnine, 2 mgm./100 ml.; – – –, from liver; . . . . . , from liver with caffeine added. Solvent, 95 per cent ethyl alcohol

With the addition of an ammonium reineckate solution the conjugate acid, BH^+, thus formed will react with the $[Cr(NH_3)_2(SCN)_4]^-$ ions arising out of the dissociation of ammonium reineckate and form the relatively insoluble reineckate complex, thus:

$$BH^+ + [Cr(NH_3)_2(SCN)_4]^- \rightarrow BH[Cr(NH_3)_2(SCN)_4] \qquad (2)$$

The equilibrium represented by equation (1) is disturbed and more conjugate acid will be formed to replace the reineckate salt as it is removed. It is obvious that in an aqueous solution the formation of the reineckate complex depends on the formation of the conjugate acid BH^+ from the base *B*.

The equilibrium between the conjugate acid and water is:

$$BH^+ + H_2O \rightleftharpoons H_3O^+ + B \qquad (3)$$

The extent to which this equilibrium will exist will depend not only on the strengths of the bases but also on the *p*H of the reaction media. The relation between these variables may be expressed by an equation of the form

$$\log \frac{[B]}{[BH^+]} = pH + pK_b - 14 \qquad (4)$$

where pK_b is the dissociation constant of the base.

In practice, the pK_b values of the bases can be obtained, and by using equation (4) the required *p*H of the reaction media can be calculated; this will either be favourable or unfavourable for the formation of a particular reineckate. Thus piperidine (pK_b 2·8) and pyridine (pK_b 8·6) can be separated in an ammonical solution of ammonium reineckate at *p*H 9. After the separation of the piperidine reineckate, acidification of the filtrate to *p*H 2 aids the formation of pyridine reineckate.

In cases where the compounds are not very soluble in an aqueous solution the reaction may be carried out by dissolving the bases in chloroform or benzene and shaking it with an aqueous solution of ammonium reineckate of the required *p*H. The separation of codeine (pK_b 6·05) and caffeine (pK_b 13) at *p*H 7 was effected in this way. Tables 1 and 2 show recoveries of piperidine and pyridine, and codeine and caffeine, respectively, from mixtures.

Table 1. Recovery of Piperidine and Pyridine from Mixture

Piperidine used (mgm.)	Pyridine used (mgm.)	Piperidine recovered (mgm.)	Pyridine recovered (mgm.)
83·0	147·3	82·6	145·0
150·8	38·7	151·2	37·3

Table 2. Recovery of Codeine and Caffeine from Mixtures

Codeine used (mgm.)	Caffeine used (mgm.)	Codeine recovered (mgm.)	Caffeine recovered (mgm.)
52·4	256·3	51·4	252·7
15·7	482·4	15·2	477·5

The separation of basic putrified materials (mainly ptomaines) from basic nitrogenous poisons from highly decomposed human liver has been effected by precipitation of the interfering substances in ammoniacal solution of *p*H 9. The purity of the recovered strychnine from highly putrified human livers using this procedure is demonstrated in Fig. 1.

A dibasic substance may be treated as a mixture of two bases since it possesses two different dissociation constants. In agreement with the reaction mechanism just discussed many of the dibasic antihistamines studied gave the di-reineckate derivatives in acid solution (*p*H 2); in water (*p*H 7) only the mono-reineckates are obtained. Equation (4) also shows that the reineckates of very weak bases can only be formed in strongly acidic media. Urea, which is reported as not forming a reineckate[1], has yielded a reineckate under these conditions.

Thus, with a knowledge of the reaction mechanism, ammonium reineckate has become a very versatile and useful reagent. It can be used for the separation of certain mixtures of bases besides forming derivatives with them which are useful for identification and determination. Molecular weights of the conjugate bases can be obtained by spectrophotometric analysis of the complexes. The free bases can also be regenerated easily by using ion-exchange resins.

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Swift Dumas Nitrogen Estimation

A novel apparatus, Fig. 1, has been designed which enables the rapid estimation of nitrogen by the principle of Dumas and Pregl[1].

The apparatus (patent applied for by Loughborough Glass Co., Ltd., Loughborough) comprises two concentric combustion tubes C_1 and C_2. The centre tube C_1 has a side-arm A passing through the outer tube C_2. C_2, having twice the diameter of C_1, is sealed at one end, and has a B.S. cone $C19$ at the other end.

When initially setting up the apparatus, a copper gauze plug N is inserted into the far end of tube C_1, followed by a layer of fine copper oxide wire CuO, 15 mm. long, which is kept in place by an asbestos plug P_1. Tube C_1 is stoppered with a rubber bung R_2, and the annular space between the tubes is packed

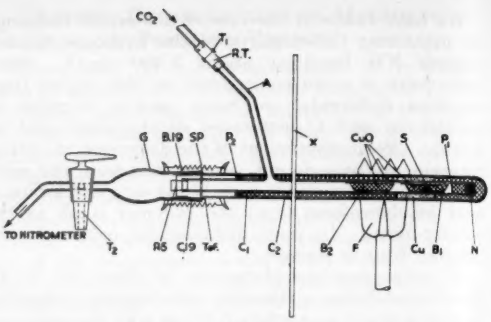

Fig. 1

with fine copper oxide for 50 mm., followed by a 30 mm. filling of copper wire and finally copper oxide wire within a distance of 50 mm. from cone $C19$. The filling of tube C_2 is kept in place by asbestos plug P_2. Side-arm A is connected to a carbon dioxide generator using a rubber tube R_T and a screw clip T_1. An asbestos sheet X is arranged over the combustion tube as in Fig. 1, and, with carbon dioxide passing through, the tubes are heated and left to cool.

Through G, which consists of a $C19$ socket and a three-way tap T_2, the apparatus is connected to the nitrometer. The cone and socket joint is sealed with a 'Teflon' sleeve *Tef*, the connexion being secured with springs.

For analysis, a sample of 3–7 mgm. is weighed into a suitable container, which is then placed in a metal boat B_1 and covered with copper oxide. With tap T_1 open, bung R_2 is removed and boat B_1 is placed in the far end of tube C_1. A second boat B_2 filled with copper oxide is placed in front of it. Bung R_2 is replaced and cone $C19$ with the 'Teflon' sleeve attached to it, connected to socket $C19$.

With tap T_1 open and tap T_2 open to the atmosphere, carbon dioxide is bubbled through for about 1 min. Tap T_2 is then opened to the nitrometer. As soon as micro-bubbles are obtained and the froth is taken off in the nitrometer, the combustion can be started.

Tap T_1 is closed and tap T_2 opened. Flame F is placed under the part of the tube containing the copper layer and slowly moved towards the end of the tube. Combustion is complete after about 5 min., when escape of bubbles into the nitrometer will stop. Tap T_2 is then closed and tap T_1 opened. T_2 is then adjusted so that bubbles escaping into the nitrometer follow each other closely. The flame is removed and the tube left to cool. After about 5 min. carbon dioxide micro-bubbles will appear again in the nitrometer. Tap T_2 is connected to the atmosphere and after the usual period the volume of nitrogen is read.

Results obtained using the procedure described are given in Table 1. More details will be published later.

Table 1

	Sample (mgm.)	Percentage nitrogen (theory)	(found)
Dinitrobenzene	4·546	16·67	16·60
	4·778		16·63
	4·694		16·71
	5·251		16·62
Azobenzene	5·060	15·37	15·30
	4·664		15·38
	3·512		15·47
	6·578		15·34

My thanks are due to Miss Y. Law and Mrs. V. Taylor for most efficient assistance; to Mr. Evans for skilful work in making this apparatus; to Mr. Lambie for encouragement throughout and to the Directors of Fisons Pest Control, Limited, for permission to publish this communication.

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A Minor Anthocyanin Pigment of Sweet Cherry

THE sweet cherry contains an anthocyanin, keracyanin, the rhamnoglucoside of cyanidin, which was first isolated by Willstätter and Zollinger[1]. Robinson and Robinson[2] indicated the presence of a cyanidin-3-monoside in the skins of small black cherries. The complete picture of the anthocyanin pigments of any one species of sweet cherry, however, has not been reported.

Recently a method of isolation of the natural anthocyanins involving silicic acid chromatography has been developed in this laboratory, and proved useful in pigment studies in sour cherries[3] and the wild strawberry[4]. The present work was undertaken to extend this method to the case of sweet cherries[5].

The crude anthocyanin pigments of sweet cherries (*Prunus avium* L., var. Windsor) were prepared by the procedure previously described for sour cherries[3]. Two pigment components of the crude anthocyanin mixture were separated from the silicic acid column, employing the upper phase of the solvent system, *n*-butanol/acetic acid/water/benzene (4 : 1 : 5 : 2 v/v) for developing and elution.

The major anthocyanin pigment, which separated as the upper band on the silicic acid column, comprised about 85 per cent of the total pigments, as compared with the 15 per cent for the minor pigment, which constituted the lower band. These proportions of the two anthocyanins were the same both in the skin and in the highly coloured flesh of the sweet cherry.

The major anthocyanin pigments, after being eluted from the silicic acid column, were crystallized from methyl alcohol containing 2 per cent hydrochloric acid in the characteristic brown-orange needle clusters of antirrhinin as isolated from sour cherries[3]. This major fraction was shown to be cyanidin-rhamnoglucoside or keracyanin by the same procedure as used for the identification of the antirrhinin from sour cherries[3].

The minor fraction of pigment was obtained as tiny red spherical crystals after elution from the silicic acid column and crystallization from methyl alcohol containing 2 per cent hydrochloric acid. On paper chromatograms this minor pigment has R_F values identical with a synthetic sample of chrysanthemin, cyanidin glucoside, in several solvents, but these numerical values were only very slightly greater than those of synthetic idaein, cyanidin galactoside, which was also used as a control. Both the minor pigment and the chrysanthemin showed identical absorption curves with maxima at 515 and 280 mμ in 0·5 per cent hydrochloric acid solutions. In 60 per cent ethanol containing 0·1 per cent hydrochloric acid, both the major and minor anthocyanin pigments of the sweet cherry showed the same maximum absorption at 535, 333 and 282 mμ.

The crystalline anthocyanidin from acid hydrolysis of the minor pigment was identified as cyanidin by the tests previously employed for the cyanidin obtained from sour cherries[3]. From the sugar solution after hydrolysis, an osazone was prepared with phenylhydrazine, with crystalline forms of glucosazone, m.p. 205° alone or admixed with authentic glucosazone. That this sugar moiety was glucose was confirmed on paper chromatograms using several of the solvent systems of Jermyn and Isherwood[6], and found to have the same R_F values as authentic glucose. Galactose, fructose, and mannose were included as controls. The minor anthocyanin pigment of the sweet cherry is therefore chrysanthemin.

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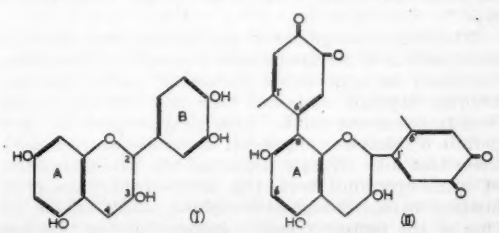
Leuco-Anthocyanins as the Possible Precursors of Tannins

ROUX has recently reported[1] that he had found leuco-anthocyanins in wattle extract and discussed biochemical aspects of these substances. The occurrence of leuco-anthocyanins in wattle and certain other tannin extracts was first referred to by Hillis in 1954[2] and further discussed by him in 1956[3]. At the same time the suggestion was put forward that leuco-anthocyanins could be the precursors of certain tannins[3,4]. Other workers also have confirmed the presence of leuco-anthocyanins in tannin extracts, and in particular in wattle extract (ref. 5 and King, H. G. C., and White, T., personal communication, May 1956).

Freudenberg[6] and others have suggested that condensed tannins are polymerized catechin (catechin I). Hathway and Seakins[7] have obtained evidence to support the view that a catechin polymer formed at *p*H 4–8 has linkages as in II. They have shown the similarity of this structure to those of the tannins from *Acacia catechu* and *Uncaria gambir*.



The leuco-anthocyanins which have been characterized possess a 3,4-flavandiol structure, and a polymerization similar to that of catechin would be expected to take place. However, it is difficult to reconcile the ease with which anthocyanidins are obtained from polymerized leuco-anthocyanins with such a structure.

A study of the amount and type of linkages in tannins has shown that those in a catechin polymer differ from those in certain tannins, and that those in the latter differ between themselves.

The method of examination involved the determination of the amount of formaldehyde which reacted with the sample at *p*H 4–5 for 30 min. at room temperature and for 3 hr. on a steam bath. We have found (unpublished work) that the catechol fragment does not react under these conditions, and thus the number of vacant reactive positions in the *A* ring can be determined. If substitution occurred in the *A* ring, its ability to react with formaldehyde would be reduced.

To avoid changes in the leuco-anthocyanins during purification crude samples of tannins were examined. They were commercial extracts of wattle bark (*Acacia mollissima*), 'Myrtan' (from wood and bark of *Eucalyptus wandoo*), mangrove cutch (from barks of *Rhizophora mucronata*, *Rhizophora apiculata* and *Bruguiera gymnorrhiza*). Kino collected from *Eucalyptus calophylla* was also examined. The tannin contents given in Table 1 have been determined by reaction with formaldehyde under acid conditions[8] and the formaldehyde uptake has been expressed on the basis of these figures.

Table 1. Properties of Catechins and Tannins

Sample	Tannin (per cent)	Formaldehyde uptake		Anthocyanidin produced*	
		30 min. at 20° C. gm./gm. tannin	3 hr. at 100° C. gm./gm. tannin	Per cent	Calculated† as :
(+)-Catechin	116	0·146	0·082	—	—
(+)-Catechin polymer‡	103	0·062	0·048	—	—
Kino	99·6	0·041	0·059	12·1	cyanidin and pelargonidin (1 : 1)
'Myrtan'	67·7	0·011	0·043	10·5	cyanidin and pelargonidin (1 : 1)
Cutch	77·7	0·027	0·040	9·9	cyanidin and delphinidin (1 : 1)
Wattle	94·9	0·016	0·065	7·1	cyanidin

* Produced by heating with butanol and hydrochloric acid (ref. 10).
† Using values of Schou (ref. 11).
‡ Prepared at *p*H 8 phosphate buffer (ref. 9).

From the results in Table 1 it may be calculated that 1·62 moles of formaldehyde reacted with each mole of (+)-catechin, whereas 0·61 moles reacted with each basic unit of a (+)-catechin polymer[9]. This loss of one available position on polymerization supports the view that it is of the head-to-tail type.

When chromatograms of the tannins were sprayed with vanillin-hydrochloric acid, it was found that kino contained an appreciable portion of mobile flavans, whereas 'Myrtan' contained very little and none could be detected in the cutch. This spray reagent does not permit a reliable comparison of wattle extract with these tannins. Wattle produced the lowest amount of anthocyanidin from the leuco-anthocyanins on heating with butanol–hydrochloric acid[10] (Table 1). One of the factors possibly responsible for this low yield is that different types of linkages may exist in the wattle tannin.

The greater reaction of formaldehyde (formaldehyde uptake) with kino than with the other tannins at room temperatures may be due to the higher amount of monomeric flavans. However, this formaldehyde uptake is still lower than that of catechin or catechin polymer, showing that in these conditions there are fewer reactive positions in the *A* rings. The commercial tannin extracts showed different amounts of blockage to formaldehyde in the *A* ring.

Table 1 shows that, whereas the formaldehyde uptake of catechin polymer and catechin decreased on heating, it increased in the case of tannin extracts, the increase being lowest for kino and highest for wattle. The above decrease was found to be due to expulsion of formaldehyde during polymerization (unpublished work). While this reaction may occur in the tannins, the overall effect is an increase in formaldehyde uptake on heating. Some of this behaviour could be due to a difference in reactivity under different conditions. On the other hand, the results are consistent with the view that a labile grouping is split off from the *A* ring under the conditions of the test. It is known that polymeric flavandiols break down readily when heated with alcoholic hydrochloric acid. Thus, it is possible that they are also broken down under the conditions of the formaldehyde reaction, liberating further positions for attack by formaldehyde. If polymeric flavandiols are linked differently in the various tannins so that some are broken down to the monomers more readily than others, then different formaldehyde uptakes could be obtained on heating (Table 1).

In addition to the different amounts of reaction with formaldehyde, catechin polymer did not react with vanillin/hydrochloric acid, whereas the tannins examined did so. Consequently, the grouping that is linked to the *A* ring must be of such a nature that it does not de-activate this ring[10] for the vanillin/hydrochloric acid reagent.

Further distinction between catechin polymer and the tannins was shown by spectral data. At *p*H 8 catechin polymer showed a maximum at 410 mμ (see also ref. 9). This maximum was not observed in freshly prepared solutions of the tannins at *p*H 8, but on standing, cutch only showed maxima at 410 and 500 mμ. Thus, cutch contains a structure capable of change to one showing a spectrum similar to that of *Acacia catechu* and *Uncaria gambir* tannins[7].

These results will be discussed fully elsewhere.

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Characterization of Ficin

THE general attributes of ficin and its enzymic action have not been thoroughly described in the literature. The work of Winnick *et al.*[1] showed conclusively that ficin was a sulphydryl enzyme. More recently the kinetics of ficin on arginine derivatives have been studied[2]. In the present communication its electrophoretic and ultracentrifugal behaviour are described. The optimal conditions for activity and stability were also established. In further characterization of its properties, the effect of acetylation of its free amino-groups on the activity and stability was studied.

To prepare a sample suitable for electrophoretic and ultracentrifugal studies a procedure was adopted to permit proper equilibration of the crystalline suspension of enzyme with the buffers desired and to ensure a depressed oxygen level. 25 mgm. of ficin was dissolved in 3 ml. of buffer and inserted into a 'Cellophane' bag which was slipped over a test tube to form a thin layer of solution around the tube. The sample was then immersed in 400 ml. of buffer kept cold in an ice-bath, through which nitrogen had been passed for 15 min. Rapid dialysis was carried out by shaking the entire apparatus for 2 hr. while the gas was continuously bubbled through the buffer.

The electrophoretic and ultracentrifugal patterns indicated the relative homogeneity of the protein. The sedimentation constant was calculated to be 2·66 S. and was found not to vary with the *p*H. The isoelectric point is at *p*H 9·0.

The *p*H optimum for proteolytic activity of ficin, employing the method of Kunitz but with 0·01 *M* cysteine present, was determined. A broad optimum from *p*H 6·5 to 9·5 was observed.

Acetylation of the free amino-groups of ficin was carried out in a solution half saturated with sodium acetate and 0·15 *M* potassium cyanide. 100 mgm. of ficin dissolved in 5 ml. of this medium, cooled in an ice-bath, was acetylated with 0·6 ml. of acetic anhydride by adding aliquots of 0·1 ml. over a 1 hr. period. The *p*H was maintained above 4·0 throughout the reaction. 40 per cent of the amino-groups were acetylated as determined by ninhydrin colorimetric analysis. Neither the activity nor *p*H optimum was altered by this modification. From the fact that acetylation with acetic anhydride, which is specific for amino-groups[3], did not impair enzymic activity it may be concluded that most of the amino-groups are not essential.

The effect of varied concentrations of three reducing agents, cysteine, potassium cyanide and dimercaptopropanol, on the activity of ficin was also investigated. The results are summarized in Fig. 1. Activation by cysteine was constant between 5×10^{-3} and 5×10^{-2} *M* concentrations, likewise the activation by dimercaptopropanol remained constant but at a lower level. In contrast, the activity of the enzyme increases with rising concentrations of potassium cyanide to a maximum at 0·075 *M*.

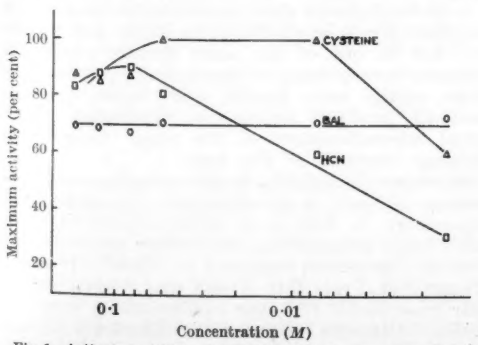

Fig. 1. Activators of ficin. 0·1 *M* phosphate buffer was employed

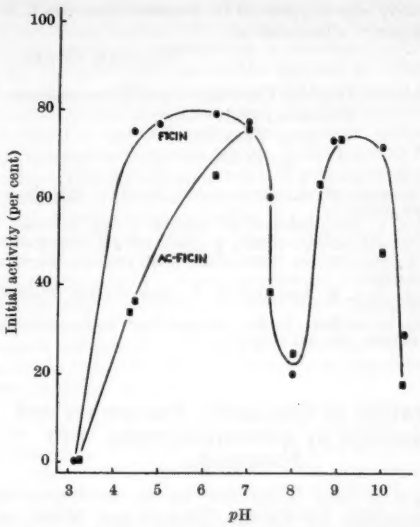

Fig. 2. *p*H stability of ficin and acetylated ficin. The enzymes were incubated for 2 hr. at 50° C. The 0·05 *M* buffers employed are as follows: glycine, *p*H 2·5–3·5; sodium acetate, *p*H 4·0–6·0; sodium phosphate, *p*H 6·0–8·0; sodium borate, *p*H 8·0–9·5; glycine, *p*H 9·5–10·5. 0·01 *M* cysteine was included in each buffer

The stability of ficin was investigated in 0·05 *M* buffers containing 0·01 *M* cysteine at a concentration of 0·33 mgm. per ml. An incubation period of 2 hr. at 50° C. was employed in these studies. The enzyme is stable between *p*H 4·5 and 9·5 except for a narrow zone around *p*H 8·0. These results are recorded in Fig. 2. The acetylated enzyme, which is also depicted in the same figure, presents similar stability properties in neutral or alkaline media, but in acid media it is considerably less stable than the native enzyme. The minimum exhibited by ficin is dependent, most probably, on the rate of auto-oxidation of cysteine rather than a property of the enzyme itself. This zone of instability near *p*H 8·0 can be effectively altered by increasing the concentration of cysteine. It was found that 0·025 *M* cysteine was optimal for stabilizing the enzyme at *p*H 8·0.

Employing 0·025 *M* cysteine, the *p*H stability was redetermined. The enzyme exhibited a more normal optimal stability near *p*H 7·5. Thus the true stability of the enzyme is that which is observed under these conditions of higher cysteine concentration.

The loss of stability in the acid-range upon acetylation of enzymes was previously noted with other enzymes[4–6]. It has been suggested that the groups governing the stability of the native enzyme do not necessarily influence the stability of the acetylated form. The principle that a few key groups are responsible for preservation of the characteristic native secondary structure of proteins can be maintained, provided it is extended to include the possibility that by redistributing overall charges through acetylation other groups may assume this function.

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Separation of Chalcones, Flavanones and Flavonols by Chromatography with Polyamide

Polyamide had been used as an adsorbent in chromatography by Carelli, Liquori and Mele[1], as well as by Grassmann, Hoermann and Hartl[2], for the separation of phenols. Recently, Hoerhammer, Wagner and Leeb[3] used it for the separation of flavones and their glycosides, and for the quantitative removal of tanning substances from plant extracts containing flavones.

The complete conversion of chalcones into flavanones is very unusual, and separation by recrystallization is often difficult. It is possible, however, to fractionate chalcones and flavones on a column with powdered polyamide; the chalcone is retained in the upper part of the column, whereas the flavanone is readily eluted with water, aqueous alcohol or aqueous acetone respectively. By this means the simple chalcones can be separated from the corresponding isomeric flavanones; for example, 2-hydroxy-, 4′-hydroxy-, 4′-methoxy-, 3′-methoxy-4′-hydroxy-flavanones. The chalcone that remains in the upper part of the column can be eluted with absolute methanol. If plant extracts are used that contain both chalcones and flavanones, for example butin and butein, the best separation is obtained by aqueous alcohol mixtures. The plant extracts are either dried on, or triturated with, polyamide and the combination placed on a polyamide column. The flavanone is first eluted with mixtures of water and alcohols or ketones, and then the chalcone with the pure solvent. The concentration of the solvent mixtures used for the separation depends on the substance, and is found by the usual alteration of the elution medium in sorption chromatography.

It is often not easy to isolate pure flavanoids, as distinct from the fraction containing these substances, and the process often involves considerable losses of substance[4]. Chromatographic procedures are especially important in this connexion but all the usual substances known as adsorbents cannot be used for the separation of flavonols, especially those that adsorb these compounds irreversibly. Polyamide is an adsorbent that has not hitherto been examined in detail in this respect.

In further work on the hydroxy phenyl-benzo-γ-pyrones it was found that separation of flavonol and flavanonol was possible on a column of polyamide. For these investigations a crude sample of dihydroquercetin that contained a small amount of quercetin was used. The extract obtained from the crude flavanonol by means of diethyl ether was dried on powdered polyamide, and then put on a column containing the same adsorbent made up in water. By elution with aqueous ethanol (20–30 per cent of ethanol) the dihydroquercetin was first obtained and then, by elution with pure ethanol, the quercetin. Both of them were paper-chromatographically pure.

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Reactivity of Sulphonyloxyl Groups with Halides in the Carbohydrate Series

It has recently been shown[1] that refluxing 6-O-mesyl-1 : 2-3 : 4-di-O-*iso*propylidene-D-galactopyranose[2] (i) and 3 : 5-O-benzylidene-6-O-mesyl-1 : 2-O-*iso*propylidene-D-glucofuranose[3] (ii) with potassium fluoride in ethanediol, yields the 6-deoxy-6-fluoro-D-galactopyranose and D-glucofuranose derivatives, respectively. A notable difference in the reactivity of the two sugars towards potassium fluoride was found (Table 1).

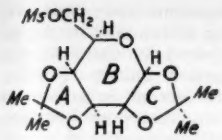
Table 1

Compound	Temperature (°C.)	Time (min.)	Per cent yield of fluorosugar
6-O-Mesyl*-D-galactose derivative (i)	200	75	60
6-O-Mesyl-D-glucose derivative (ii)	200	2·5	75

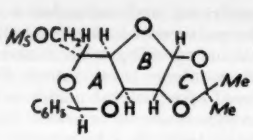
* Mesyl = $-SO_2CH_3$

A similar qualitative difference in the reactivity of terminal sulphonyloxyl groups of the above D-galactose and D-glucose derivatives has also been observed in the case of iodine exchange reactions, using sodium iodide in acetone (review by Tipson[4]). It is to be expected that the difference in size (and electronic properties) between the iodide and fluoride ions will be one of the many factors which will influence the reactivity of the sulphonyloxyl groups. These results with fluorine and iodine exchange reactions, however, suggest the importance of the actual stereochemistry of the sugar molecule in exchange reactions of this type.

Structures (I) and (II), representing the D-galactopyranes (i) and D-glucofuranose (ii) derivatives, respectively, in their most stable *cis*-anti-*cis* (rings *ABC*) steric arrangement, are written in accordance with the convention suggested by Mills[5a]. It is significant that if ring *B* in (I) and ring *A* in (II) retain their most stable *C*1 chair conformation[6], then the residue —CH_2OMs is equatorially disposed in (I) and axial in (II).



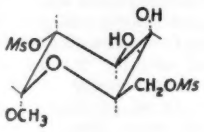
(I)



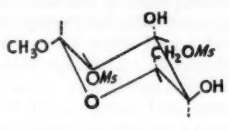
(II)

It has already been pointed out by Mills[5b] that in iodine exchange reactions, terminal axially disposed sulphonyloxyl groups in *cis* bicyclic diacetals are more readily replaced than equatorial. The fluorine and iodine exchange reactions on (I) and (II) support this view.

It is interesting to note that when (I) is converted to methyl 2 : 6-di-O-mesyl-D-galactopyranoside (III) iodine exchange at C_6 under identical conditions now proceeds much more readily[7]. This may be due to the fact that the removal of the *iso*propylidene groups from (I) now permits the —CH_2OMs residue of (III, *C*1) to take up an axial disposition (III, 1*C*).



*C*1



1*C*

(III)

Further experiments with carbohydrates of known and fixed conformation are in progress designed (*a*) to establish by kinetic studies the influence of steric factors and size of attacking anion on the reactivity of sulphonyloxyl groups, and (*b*) to provide a means of distinguishing between axial and equatorial substituents in carbohydrates where conformation is uncertain.

This work is being supported by a grant from the University of California, Los Angeles, and a grant to Dr. E. M. Gal from the National Institute of Neurological Diseases and Blindness, U.S. Public Health Services (No. *B*-1159).

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Itaconicoxidase: an Enzyme from an Ultra-Violet-induced Mutant of *Aspergillus terreus*

During an investigation of the metabolic patterns of ultra-violet-induced mutants in *Aspergillus terreus* Thom[1,2], a soluble, cell-free enzymatic extract, that can oxidize itaconic acid to itatartaric acid, was prepared from the mycelium of a morphologically altered variant.

Cell-free extracts are obtained from surface cultures of the organisms, grown at 28° C., on 100 ml. portions of the medium described in a previous publication[2]. The mycelial pad covers the surface 4–6 days after inoculation. The original culture medium is replaced by 100 ml. distilled water, and the mycelium is allowed to stand in this water for 1 hr. at room temperature. All subsequent operations are carried out in the cold-room. The mycelium is washed with several portions of ice-cold water and is then ground in a mortar with 3 ml. of 0·3 *M* citrate buffer, *p*H 4·16, in the presence of about 1·5 gm. of glass beads. The paste is diluted with 7 ml. of citrate buffer and centrifuged at 1,500*g* for 20 min. at 0° C. The supernatant is passed through a filter paper, yielding an opalescent solution with a slight orange–tan colour. Such solutions have *p*H values between 4·1 and 4·2, and contain some itaconic acid, or itatartaric acid released from the cells during the grinding process.

The enzymatic oxidation reaction of itaconic acid was followed manometrically by standard Warburg techniques in air at 37° C. The total volume was 3·2 ml., containing 1·5 ml. of 0·25 per cent itaconic acid substrate in citrate buffer; 1·5 ml. cell-free enzyme in citrate buffer, *p*H 4·16; and 0·2 ml. of 2 *N* potassium hydroxide were in the centre well.

The other details of the manometric methods and culture conditions, as well as chemical analytical methods used for sugars and acids, the irradiation techniques for the production of the mutations, and the selective work are described in preliminary studies[3].

Typical results of manometric studies of the oxidation of itaconic acid by enzymes prepared from the mycelium of the parent strain (*A*) and of the mutant (*B*), as well as oxygen uptake by enzyme controls in the absence of substrate (endogenous respiration, *E*) are shown in Fig. 1. The enzymatic oxidation of itaconic acid expressed as percentage of the original quantity is shown in Fig. 2. The enzyme obtained from the mutant, which is most active over the *p*H range 4·0–4·2, readily oxidizes itaconic acid forming approximately stoichiometric amounts of itatartaric acid. On the other hand, the cell-free extracts prepared from the unirradiated parent strain did not oxidize itaconate.

The dependence of oxygen consumption on age, concentration, *p*H, inhibitors and oxygen pressure

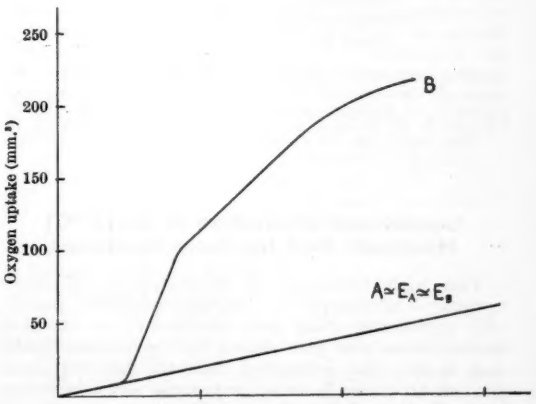

Fig. 1. For key see text

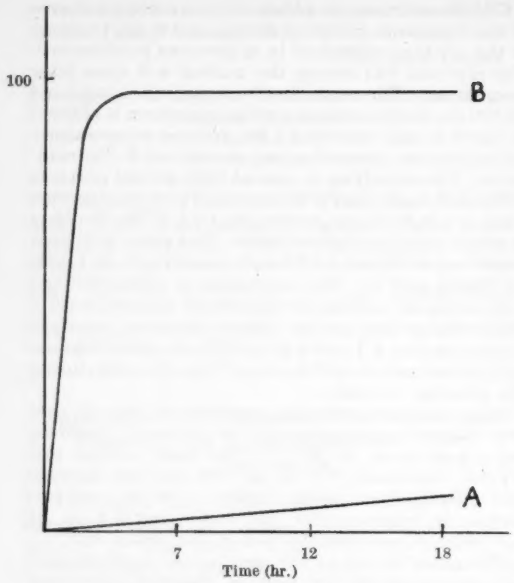

Fig. 2. For key see text

was tested. The oxygen consumption was found to be largely independent of the itaconic acid concentration, and the rate of oxygen uptake decreased with time. The course of oxygen uptake can be divided into two parts, an initial rapid phase, and a second, very slow phase. As the *p*H is decreased, the rate of itaconic oxidation increases, the oxygen consumption at *p*H 4·2 being about twice that at *p*H 7. Cyanide, hydroxylamine and iodoacetic acid, even at concentrations of 0·01 *M*, have no effect on the oxidation of itaconic acid. Dialysis does not decrease the activity.

The rate of oxidation is proportional to the oxygen pressure, the increase on changing from air to oxygen being generally 100–200 per cent.

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Uptake and Utilization of DL-[2-^{14}C] Mevalonic Acid by *Hevea brasiliensis*

THE high efficiency of incorporation of DL-3-hydroxy-3-methyl-[2-^{14}C] pentano-5-lactone (DL-[2-^{14}C] mevalonic acid) into cholesterol in rat-liver homogenates was first shown by Tavormina, Gibbs and Huff[1], who concluded that the lactone could provide an active 5-carbon isoprenoid unit which was used in direct biosynthesis. It was further found that this unit was derived from the lactone by elimination of C_1 as carbon dioxide[2]. These results have been confirmed and extended to squalene biosynthesis[3–5], the pattern of isotopic labelling determined by degradation agreeing with that required for the synthesis of squalene from intact 5-carbon units[3]. Further proof of a general utility of the lactone in isoprenoid biosynthesis is shown by its incorporation into β-carotene in *Phycomyces blakesleeanus* and carrot slices[4], and in *Mucor hiemalis*[2]. The present communication describes briefly the further extension of this field to the uptake and utilization of DL-[2-^{14}C] mevalonic acid by a rubber-producing plant, *Hevea brasiliensis*.

The plants used were 2-year old seedlings, grown in a greenhouse. They were in active growth, and carried large healthy leaves. The uptake of [2-^{14}C] mevalonic acid was best carried out by applying a solution in neutral phosphate buffer to the upper surfaces of leaves, or to lightly scraped areas of the main-stem. To aid uniform leaf application, a trace of wetting-agent ('Tween') was added to the solution. Hypodermic injection was unsuccessful, even when applied to the soft upper stem tissues.

Absorption of the lactone by the leaves was rapid, the count-rates of the upper and lower surfaces being about equal after 5 hr., thereafter remaining almost unchanged. Translocation of activity through the plant did not occur, even to neighbouring untreated leaves, and none could be induced by tapping the plant at several points along the stem. After two days the active leaves were harvested, washed thoroughly with water, ground with sand, and extracted with warm water. All the activity was water-soluble (total count-rate 12,750 c.p.m.), and re-extraction of the aqueous solution with petroleum-ether resulted in the transfer of 52 per cent of its activity. It was shown by paper chromatography that all the activity remaining in the aqueous solution was due to unchanged mevalonic acid. The petroleum-ether extract was chromatographed on a column of 'Celite', and eluted with the same solvent. At least four coloured components were resolved, of which a yellow pigment, which ran with the solvent front as a diffuse band, was completely separated from the remaining bands. This first fraction contained 96 per cent of the activity applied to the column. The residual activity on the column was eluted in the same position as a control addition of [2-^{14}C] mevalonic acid.

The absorption spectra of the active pigment in several solvents agreed closely with those known for α-carotene, and this identity was confirmed by the dextro-rotation shown by the pigment solutions. If it is assumed that only one of the enantiomorphs is physiologically active, the above results show that virtually all the available mevalonic acid absorbed by the leaves was converted to α-carotene.

Application of mevalonic acid to the stem was designed to show whether its efficient use for carotene synthesis in leaves was masking a possible alternative use for rubber synthesis. Again, absorption was rapid, activity moving down the stem from the point of application to appear in the lower newly formed side-shoots and leaves after ½ hr. No activity could be detected moving up the stem.

After three days, leaf activity accounted for about 50 per cent of the total (5×10^5 c.p.m.) applied to the stem, and was completely soluble in water. In this case, however, no activity had been incorporated into the carotene fraction, and only a small amount was due to unchanged mevalonic acid. Paper chromatography with a solvent system of *n*-butanol/water/90 per cent formic acid (10 : 10 : 3 by volume) showed

most of the leaf activity to be associated with a single component (R_F 0·12), which probably possessed more polar groups than mevalonic acid (R_F 0·72).

The remainder of the plant, namely, the main stem and side-shoots, was reduced to a fine pulp, and extracted successively with water, ethanol, acetone, ether, and benzene. About 30 per cent of the applied activity was recovered in the aqueous extract, and 20 per cent in the ethanol extract. All other extracts and residues were inactive.

The component with R_F value 0·12 appeared in both aqueous and ethanolic solutions, the latter also containing an active substance (R_F 0·93) which was probably a carboxylic acid.

It may be concluded that, under the conditions of these experiments, mevalonic acid was not incorporated into rubber in *Hevea*, and that it was converted in the stem to products unable to enter the pathway of carotene synthesis which can operate with high efficiency in the leaves.

Fuller details of this work will be published elsewhere at a later date.

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Action of Testosterone Propionate and 4-Chlorotestosterone Acetate on Protein Synthesis *in vitro*

The protein anabolic activity of testosterone propionate is well established[1]. It has been shown that an increase in the rate of uptake of labelled glycine occurs in slices of kidney from mice treated with this substance[2]. Incorporation of labelled glycine into liver proteins does not seem to be affected.

As the action of testosterone propionate on protein synthesis might depend on the condition of the tissue, we have studied the incorporation of glycine-1-^{14}C into slices of resting and regenerating livers from animals treated with testosterone propionate.

The effects of a new compound, 4-chlorotestosterone acetate, recently synthesized by Camerino, Patelli and Vercellone[3], have also been studied. This substance possesses powerful myotrophic and weak androgenic activity, as measured by increase in weight of levator ani muscle and ventral prostate gland, respectively[4]. Having greater anabolic and lesser androgenic effects than testosterone propionate similarly administered, it may have considerable therapeutic value as an anabolic agent.

Castrated male rats, weighing 200–250 gm., were used throughout our experiments. They were treated subcutaneously with 2 mgm. of testosterone propionate or 4-chlorotestosterone acetate once a day for three days. The first injection was done 2 hr. after partial hepatectomy in the case of the animals with regenerating liver. At the end of the treatment (72 hr. from the start, 74 hr. from hepatectomy) the rats were killed by decapitation, the livers quickly removed and sliced by hand with a razor. The slices were then placed into the main chamber of a Warburg flask containing 3·0 ml. of phosphate-saline[5]. After 10 min. equilibration, the labelling reaction was started by transferring 2 μc. (3·4 μmoles) of glycine-1-^{14}C in a volume of 0·4 ml. from the side-arm into the main chamber of the flask: this was within 30 min. of the death of the animal. The labelling reaction lasted 1 hr.; the experimental conditions and counting techniques were the same as described by Bernelli-Zazzera and Guidotti[5].

The results are shown in Table 1.

Table 1. Incorporation of Glycine-1-^{14}C into the Proteins of Slices of Resting or Regenerating Livers from Animals treated with Testosterone Propionate or 4-Chlorotestosterone Acetate

Counts/min./mgm. of protein. Means of five experiments ± S.E.

	Resting	Regenerating
Controls	165·3 ± 14·0	257·8 ± 40·2
Testosterone propionate treated	152·9 ± 10·8	326·0 ± 36·1
4-Chlorotestosterone acetate treated	165·1 ± 21·6	409·6 ± 47·9

Sum of squares between experiments, within conditions = 73,274·75 (8 d.f.).

The Bartlett test is not significant. Therefore we made an analysis of variance, considering that three rats (control and one treated with each drug) were used throughout one experiment. It was possible to conclude that: (1) slices of regenerating livers incorporate glycine at a rate significantly higher than that of resting livers ($P < 0·01$). (2) Slices from rats treated with testosterone propionate or 4-chlorotestosterone acetate incorporate glycine at a rate significantly higher than untreated controls ($P < 0·05$). (3) There is no difference between the effects of the two treatments ($P > 0·05$). (4) The effects of the treatments largely depend on the state of the tissue ($P < 0·05$); this can be seen from Table 1, where the treatments are effective in enhancing glycine incorporation into liver proteins only in the case of regenerating livers, when the restoration of cell structures takes place and the rate of protein synthesis reaches its maximum. Therefore, the hypothesis that testosterone propionate and 4-chlorotestosterone acetate are mainly active on the synthesis of cell proteins is not inconsistent with our results.

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Effect of Atmospheric Contaminants on Paper Chromatography of Urinary Indole and Phenol Acids

Samples of human urine have been examined for indole and phenol acids as a matter of routine in this laboratory for several months; the methods used for extraction and two-dimensional paper chromatography were described earlier[1,2]. The chromatograms were satisfactory until certain days in April, when the normally pink and purple spots given by indoles with *p*-dimethylaminobenzaldehyde showed a diminished colour intensity and brown discoloration. The colours became so weak in May that indole spots could not be seen with certainty. Unlike the indoles, the phenolic acids gave colours which were normal immediately after spraying with diazotized sulphanilic acid, but which faded and were discoloured afterwards to an increasing extent. No improvement was obtained when the components of the chromatographic solvents were purified extensively.

The occurrence of chromatographic difficulties appeared to coincide with an increased level of atmospheric contaminants. The eye-irritating haze known as 'smog', develops frequently in the Los Angeles area where topography and meteorological conditions favour the formation of a large stagnant air mass and the accumulation of fumes from numerous industries and other sources. 'Smog' contains ozone, organic peroxides, nitrogen oxides and other components[3]. The 'smog' level was low early in 1958 because of unusually heavy rains, but became noticeable in April and marked in May; an atmospheric ozone content of 0·35 parts per million was reported in Pasadena on May 15.

Several other features suggested that oxidative atmospheric contaminants were responsible for the destruction of indole spots on chromatograms. Ozone has been used preparatively to rupture the pyrrole ring of indole compounds[4]. The instability of ozone and organic peroxides in alkaline media is well known; on the other hand, ozonization is frequently carried out in acetic acid. These properties may explain why one-dimensional chromatograms developed in *iso*propyl alcohol/ammonia/water (8 : 1 : 1) furnished indole spots with almost normal colours with *p*-dimethylaminobenzaldehyde at a time when those run in benzene/propionic acid/water (100 : 70 : 5) did not.

A pertinent counterpart to these observations was provided by a report concerning the adverse effects of 'smog' on the response of plants to indoleacetic acid[5]; the 'smog' was eliminated by filtering the air through charcoal[6]. This approach also afforded a solution to the present problem. Two sets of two-dimensional chromatograms were prepared from four different urine extracts. One set was dried in the hood as usual; *p*-dimethylaminobenzaldehyde revealed faint brown (instead of blue) spots of only 5-hydroxyindoleacetic acid. The second set was dried in empty chromatography jars, after each solvent and after spraying, by a current of air which had passed through a 2-l. aspirator bottle filled with 8–12 mesh activated charcoal; 14 indole spots with normal colours were observed on this set of chromatograms. A charcoal unit large enough to ensure a 'smog'-free room is being designed.

The formation of 'smog' is a problem of increasing importance in densely populated and heavily industrialized cities. These findings, therefore, may be of interest to investigators working in such areas with small amounts of compounds or biological systems which are sensitive to the action of atmospheric contaminants.

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Inhibitory Action of Ethyl Mercaptan on Intracellular Tubercle Bacilli

We have previously described the anti-tuberculous activity in mice of ethyl mercaptan and its derivatives[1,2]. The marked activity which was observed did not seem to correspond with the slight bacteriostatic action of ethyl mercaptan against *Mycobacterium tuberculosis in vitro*. Since the bacilli in tuberculosis are intracellular, we decided to test the effect of ethyl mercaptan against bacilli so located. For this purpose we used the method described by Suter[3] in which tubercle bacilli are grown *in vitro* within monocytes derived from peritoneal exudates of normal guinea pigs. Because of the high volatility of ethyl mercaptan it was necessary to modify Suter's method of incubating the cover-slips on which the monocytes and bacteria were attached. The tubes containing the cover-slips were sealed by heat before they were incubated, at an angle of 30° from the vertical, without rotation. This modification still allowed good growth of the bacteria and isoniazid was highly effective (Table 1).

Four cover-slip preparations from each treatment were stained at intervals of 1, 3, 5 and 7 days and 200 monocytes were examined on each slide and classified as containing 0, 1–10, 11–20, and more than 20 bacteria respectively. The activity of an inhibitory compound can be seen most conveniently from the percentage of cells containing more than 20 bacteria in the 7-day cultures.

Ethyl mercaptan was inhibitory at concentrations of the order of 10 μgm./ml., methyl mercaptan was ineffective; indeed it partially antagonized the effect of ethyl mercaptan. This is in accordance with our observations in mice[2]. The specificity of this effect was demonstrated by the failure of ethyl mercaptan to inhibit the intracellular growth of a strain of bacilli which had been made resistant to treatment with thiol esters in mice[2] (Table 1). Several potential metabolites of ethyl mercaptan, including methyl ethyl sulphide, methyl ethyl sulphone, β-hydroxyethyl methyl sulphide and β-hydroxyethyl methylsulphone were inactive, whereas the diethyl disulphide was active.

Table 1

Treatment or conditions of growth	Percentage of monocytes containing more than 20 bacteria at 7 days
Open tubes	86
Sealed tubes	97
Sealed tubes with isoniazid 0·1 μgm./ml.	25
Ethyl mercaptan 100 μgm./ml.	9
Ethyl mercaptan 10 μgm./ml.	18
Ethyl mercaptan 1 μgm./ml.	35
Untreated controls	35
Antagonism	
Ethyl mercaptan 100 μgm./ml.	12
Methyl mercaptan 100 μgm./ml.	40
Ethyl mercaptan 100 μgm./ml. + methyl mercaptan 100 μgm./ml.	19
Untreated controls	34
Strain resistant to ethyl thiol esters (ref. 2)	
Ethyl mercaptan 100 μgm./ml.	83
Isoniazid 1 μgm./ml.	5
Untreated controls	80
Normal sensitive strains	
Ethyl mercaptan 100 μgm./ml.	25
Untreated controls	68
Human monocytes	
Ethyl mercaptan 100 μgm./ml.	10
Untreated controls	63

The inhibitory action of ethyl mercaptan was also demonstrated using human monocytes obtained from a patient suffering from monocytic leukæmia (Table 1).

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A Simple Method for making Starch-gel Electrophoretic Strips Transparent

Starch-gel electrophoresis, as introduced by Smithies[1], has proved superior to both free and filter-paper electrophoresis for separating protein mixtures[2–6]. However, the opacity of the strips excludes the possibility of direct quantitative evaluation of separated proteins, though this is possible with the other two electrophoretic methods. This problem was successfully solved recently by Fine and Waszczenko[7], who prepared starch-gel suitable for direct quantitative evaluation by transmission photometry. A simpler and quicker method for obtaining transparency of the strips is presented here.

At the end of the run, the standard $\frac{1}{4}$-in. thick starch-gel strip is transferred into a $\frac{3}{16}$-in. deep slicer of the type described by Smithies[1]. This is then covered with a $\frac{1}{16}$-in. deep slicer, and the cutting is performed lengthwise between these two. In this way the upper portion of the strip is removed and then discarded. The rest of the strip is transferred into a $\frac{1}{8}$-in. deep slicer, then covered and sliced as above except that this slice is transferred to the staining dish. Similarly, another slice is cut in a $\frac{1}{16}$-in. deep slicer and is also transferred to the staining dish. In this way two $\frac{1}{16}$-in. strips of the central portion of the original strip are obtained.

Staining the strips is carried out in a saturated solution of buffalo black in a methyl alcohol/water/acetic acid (50 : 50 : 1) solution for a period of 2 min. The same solution, but without the dye, is used for removal of the background stain, which occurs after the strips have been left in the washing solution for 12–16 hr. Strips are then transferred to a tray containing a 10 per cent solution of acetic acid and boiled for 30 sec., after which the whole is cooled to room temperature. Such a procedure renders stained strips transparent. Strips should not be removed from the acetic acid solution before cooling since they are very fragile. Transparency of the strips treated thus is greater than that of freshly formed gels, while their consistency is of the same order.

It is worth mentioning that certain protein fractions which are present in minute amounts are more readily visible after the strips are made transparent.

In order to evaluate these strips photometrically, they are transferred into a $\frac{1}{16}$-in. deep slicer and covered by a 'Plexiglass' strip. Care should be taken not to trap any air bubbles below or above the strip. This can be achieved by carefully laying the strip on the slicer and delivering a sufficient amount of acetic acid solution around and on the top of the starch-gel strip prior to its covering.

If one wishes to preserve starch-gel strips for future reference, one should transfer them back to the methyl alcohol/water/acetic acid solution, in which they solidify after resuming their original opacity.

I wish to acknowledge the help and assistance of Dr. O. Smithies in introducing the starch-gel electrophoresis method in this laboratory.

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Thermal Decomposition of Antibiotics

Previous work on the thermal decomposition of the tetracycline series of antibiotics in which aureomycin, terramycin and tetracycline were studied[1] has been extended to include erythromycin, leucomycin base, oleandomycin phosphate and carbomycin.

The thermograms were obtained by use of an automatic recording thermobalance[2], using the same procedure as before[1]. The antibiotics were obtained

as commercial samples and were not further purified before pyrolysis.

The thermograms of the four antibiotics are given in Fig. 1. Although a detailed analysis of the decomposition mechanism is difficult to make, the loss of bound water is readily detected. Any further interpretation of the decomposition process is difficult to do because of the complex structure of the antibiotics and also because the pyrolysis was carried out in air.

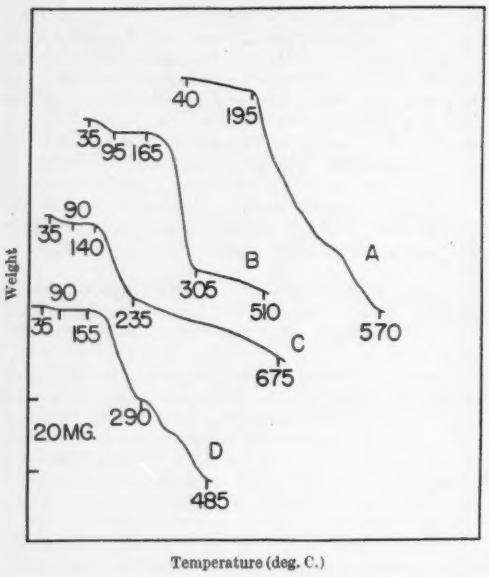

Fig. 1. Thermal decomposition of antibiotics: *A*, carbomycin; *B*, erythromycin; *C*, oleandomycin phosphate; *D*, leucomycin base

Erythromycin. The first loss in weight for this antibiotic was observed at about 35° C. After this initial loss in weight, a horizontal weight-level was found extending from 95° to 165° C. This first loss in weight is thought to be due to the evolution of loosely bound water. Previous studies have shown that this water is completely lost at 55° C. under reduced pressures[2]. Beyond 165° C. the antibiotic decomposed rapidly, leaving a small amount of carbonaceous matter (less than 10 per cent) in the thermobalance pan at 510° C.

Carbomycin. The antibiotic lost weight continuously from 40° to 570° C. Starting at 40° C., the loss in weight was quite slow until 195° C. was reached, when it began to decompose rapidly. As with erythromycin, some carbonaceous matter remained at 570° C.

Oleandomycin phosphate. The first loss in weight was observed at 35° C. After this initial loss, a nearly horizontal weight-level was observed from 90° to about 145° C. Beyond 145° C. the antibiotic decomposed rapidly, giving a break in the curve at 235° C., then further decomposition up to 675° C. Since this was a phosphate salt, the phosphate was left behind with the unoxidized carbonaceous matter.

Leucomycin base. After an initial loss in weight beginning at 35° C., a horizontal weight-level was observed from 90° to 155° C. Beyond 155° C., the antibiotic decomposed rapidly, giving a break in the curve at 290°. Further decomposition then occurred above 290° C. to give a small amount of carbonaceous matter at 485° C.

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Separation and Determination of the Dissociation Constants of Chelidonine and Protopine by Paper Chromatography

In connexion with the chromatographic separation of alkaloids of *Chelidonium maius* L.[1], I found alkaloids giving rather big spots with a tendency to form tails. This is connected, among other things, with the dissociation constant (K_B), the value of which influences the course of the chromatographic process, organic electrolytes having different values of K_B in the water phase and the organic phase of dissociation.

By the use of buffer solutions and a proper choice of *p*H, the degree of dissociation can be regulated and thus the distribution ratio. This also affects the shape and the rate of migration of bands.

Attempts were made at separating chelidonine and protopine by means of buffered paper. The application of the methods of Waksmundzki *et al.*[3,4], who use a slightly moistened buffered paper, reduced the duration of the chromatographic process significantly. The dissociation constants of the alkaloids which were also calculated from formulæ[5] could also be determined. Moistened paper is slightly adsorptive, so continuous counter-current extraction takes place. Whatman No. 4 paper and the apparatus described by Waksmundzki and Soczewiński[4] were used. The mobile phase was chloroform, which is a good solvent for the two alkaloids. Its low viscosity considerably shortened the time of analysis. The chromatogram was developed over 15 cm.

The initial attempts, using moistened but unbuffered paper, gave negative results: the alkaloids migrated with the solvent front ($R_F = 1$).

In buffering the paper seven liquids in the *p*H range 1–6·45 were prepared. The values were checked with a *p*H-meter.

In order to determine the optimal value of the 'moistening index',

$$W = \left(\frac{\text{weight of moistened paper}}{\text{weight of dry paper}}\right)$$

the studies were carried out on paper buffered at *p*H 2. The effect of moistening the paper on the behaviour of the alkaloids and the duration of the chromatographic process were observed. The results are shown in Table 1.

It can be seen from Table 1 that 1·5 is the best value of W_z since the separation was obtained in a

Table 1. Effect of Moistening the Paper on the R_F Value and Duration of the Chromatographic Process

Alkaloid	R_F with W_z = 1	1·5	1·75
Chelidonine	0	0·53	0·57
Protopine	0	0	0
Time (hr.)	1	1	4

comparatively short time. This value was used in further work.

Table 2 illustrates the dependence of R_F on the *p*H of the aqueous phase.

As appears from Table 2, acidification makes a good separation possible. The spots were distinct after development with Dragendorff's reagent, and did not form tails.

From Fig. 1 it can be seen that the two alkaloids have different dissociation constants. Assuming that the best separation is obtained when the arithmetical mean of the two values is $\frac{1}{2}$, the optimal value for *p*H will be 2·9–3·5. In this range protopine has moved from the start line and chelidonine has not reached the front (the mean R_F value is about 0·45).

The points marked (+) were obtained experimentally, whereas those marked (O) were calculated using the equation deduced by A. Waksmundzki and E. Soczewiński[5]:

$$R_F = \frac{kr}{kr + 1 + \frac{K_B}{[OH^-]}}$$

where $[OH^-]$ is the concentration of hydroxyl ions in the aqueous phase; k is the partition coefficient of the substance under examination between the organic phase and the water phase; r is the ratio of the volume of the organic phase to that of water; and K_B is the dissociation constant of the base in question.

The experimental results served to determine the dissociation constant of the alkaloids under examination. From the difference in weight between a moistened strip and one that had been chromatographed with chloroform, the weight of chloroform in a known part of the strip was calculated. This weight was converted to a volume, and hence a value of 1·86 was obtained for r.

In order to determine k, weighed portions of the alkaloid were dissolved in chloroform and shaken with an equal volume of water. The aqueous phase was titrated with 0·05 *N* hydrochloric acid and the amount of alkaloid calculated. These results were checked by weighing the residue of alkaloid after the solvent had been evaporated from the chloroform phase. k was found to be 23·4 for chelidonine and 29·5 for protopine.

Substitution of these values of k and r in equation (1) gave $3{\cdot}8 \times 10^{-11}$ and $9{\cdot}73 \times 10^{-9}$ as the dissociation constants of chelidonine and protopine respectively.

Table 2. Dependence of R_F on the *p*H of the Buffer

*p*H	R_F of chelidonine	R_F of protopine
1·00	0·04	—
1·75	0·22	—
2·20	0·53	0·0
3·50	0·94	0·13
4·60	0·98	0·63
5·68	0·99	0·93
6·45	1·00	0·98

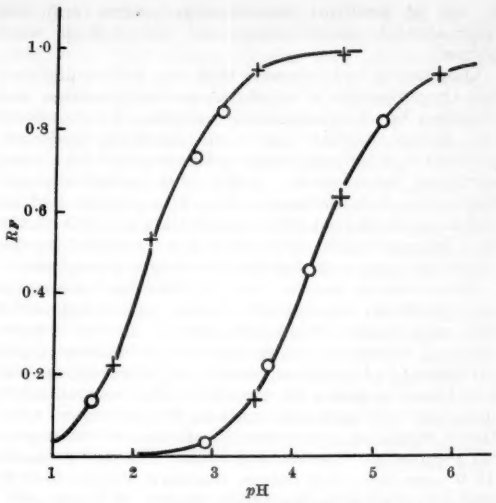

Fig. 1. Relation between R_F and *p*H for chelidonine and protopine. For key, see text

Thus the use of moistened, buffered paper permitted the separation of the substances under examination and the calculation of their dissociation constants. Because of the great differences in the partition coefficients of these alkaloids, a good separation is to be expected with Craig's countercurrent extraction method, using a buffer solution of *p*H 3·5 and chloroform.

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Riboflavin in Yellow Semen

Most specimens of bull semen are almost white, but during the course of unrelated studies[1] on spermatozoa it was observed that the semen from one particular bull was quite yellow. The colour was confined to the plasma fraction and ultra-violet light produced a green fluorescence similar to that emitted by riboflavin. Other reports[2–4] have since been noted which suggest that the yellow pigmentation of bull semen may be attributed to riboflavin, and recently eight bulls producing yellow semen have been located, thus permitting a more critical and extensive investigation. The production of yellow semen was characteristic of these bulls, which were located in scattered parts of Australia and New Zealand. All bulls producing yellow semen were of the Friesian breed, and six were the first, second or third generation descendants of one sire. The pigmentation apparently does not affect the fertility of the semen since the bulls were

in use at artificial insemination centres and the spermatozoal concentration and morphology were normal.

Qualitative tests showed that the yellow pigment had the properties of riboflavin and confirmation was obtained by chromatography of ethanol extracts of the semen using 5 per cent disodium hydrogen phosphate, *n*-butanol/acetic acid/water (4 : 1 : 5) and methanol as solvents. Light blue spots, one corresponding to lumichrome, were also present and an unknown violet spot which ran well ahead of riboflavin in *n*-butanol/acetic acid/water but remained at the origin in 5 per cent disodium hydrogen phosphate.

Quantitative aspects were studied by estimating the riboflavin content of sixteen yellow ejaculates and nine control white ejaculates. In the estimations of riboflavin[5] by conversion to lumiflavin, the preliminary chloroform extract of yellow semen was also found to give a fluorescence. This was measured quantitatively and expressed as the amount of riboflavin which on irradiation in alkaline solution gave an equivalent fluorescence. The riboflavin (mean 18·9 μgm./ml., population standard deviation 6·0) and flavin-soluble chloroform (mean 19·1 μgm./ml., standard deviation 6·3) content of the yellow samples were each about forty times that of the colourless samples and sufficient to account for the pigmentation. Both first and second yellow ejaculates obtained at any one time from individual bulls had a high flavin content which showed little variation although different samples were collected at intervals over a period of several months. It is particularly interesting that the ratio of riboflavin to flavin soluble in chloroform remained fairly constant at unity. Neither niacin[5] nor thiamin[5] values differed greatly between the groups, and the fructose levels for fourteen yellow ejaculates (mean 458 mgm. per cent, standard deviation 86) were normal.

On standing for several hours the yellow colour of the semen fades. Analysis demonstrated that this was due to the breakdown of riboflavin by sunlight rather than any metabolic activity of the spermatozoa. Chromatography showed that the lumichrome content increases at the expense of riboflavin. Ribitol, which might be expected to result from the photolysis of riboflavin, is not utilized by bull spermatozoa since the addition of 1 mgm. to 1 ml. of washed cells failed to increase the uptake of oxygen.

The results clearly establish that the yellow coloration of bull ejaculates is due to riboflavin. Yet other flavins, particularly one soluble in chloroform, are present in high concentrations even when the most rigorous precautions are taken to exclude light during collection and handling of the semen.

Blood riboflavin levels were not elevated, and other bulls under the same conditions of husbandry produced normal semen; it is thus unlikely that the colour of the semen is influenced by diet. The evidence strongly suggests that the characteristic is hereditary and that it behaves as a Mendelian dominant. In view of the blood analysis, the general tissues of these bulls are unlikely to synthesize the flavins. It seems more likely that we may be dealing with a genetic mutation in which the accessory organs, probably the seminal vesicles, have the specific ability to make, or more likely to concentrate, riboflavin and other flavins.

When added to white ejaculates at a concentration of 20 μgm./ml., riboflavin usually halved the uptake of oxygen and decreased the longevity of the spermatozoa. The possibility that this is due to the formation of hydrogen peroxide is being investigated. A full report of this work will appear elsewhere.

Thanks are due to Prof. C. W. Emmens for his interest and advice, to Mauri Bros. and Thompson for the use of laboratory facilities and to Roche Products Pty., Ltd., for gifts of chemicals. We are also indebted to the Camden Park Estate, Mr. B. Doyle, Mr. I. Martin, Mr. R. Hayman and Dr. James for making bull semen available or for checking its colour.

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Dissociation of Serological Complexes of Ovalbumin and Hæmoglobin using Aqueous Carbon Dioxide

In connexion with work on the fractionation of proteins by specific immunochemical methods, the dissociation of antigen–antibody complexes has been studied. The aim was to effect this dissociation by less drastic methods than those used hitherto for obtaining antibody[1] and if possible to isolate antigen free from antibody.

Mitz[2] showed that carbon dioxide increased the solubility of some proteins in salt-free water, and this work prompted our attempt to dissociate serological complexes with the same reagent. The procedure met with some success when applied to ovalbumin/rabbit antibody and to horse hæmoglobin/rabbit antibody systems.

Our first experiments involved the specific precipitate of ovalbumin and its antiserum prepared in the rabbit by a short term (4 weeks) immunizing procedure. The precipitate, washed free of salt, was soluble in aqueous carbon dioxide (0·035 *M*; *p*H 5·0) and partial dissociation of the complex was shown by sedimentation analysis; two boundaries (Fig. 1*a*) resolved with $S_{20,w} \simeq 5$ and 13. The slower boundary corresponded to immune globulin, the unsuppressed 'primary charge effect'[3] being mainly responsible for the low S_{20}. Control runs using rabbit γ_2-globulin[4] showed that the S_{20} was reduced from 6·56 S. (*I* 0·1, veronal buffer *p*H 8·6) to 5·1 S. in aqueous carbon dioxide. Different batches of precipitate produced similar sedimentation patterns although the S_{20} values were subject to some fluctuations. The patterns were not markedly altered by variation in the age of the complex, or in the temperature (3–36° C.) or age (2 hr. to 7 days) of the solution. In contrast, significant shifts in the equilibrium, resulting in a smaller globulin release, an increase in S_{20} of the complex and finally in precipitation, were observed when the carbon dioxide concentration was decreased to 0·01 *M* or sodium chloride was added to a concentration of about 9×10^{-3} *M*.

Free antibody (15–25 per cent) was isolated from this system by differential ultracentrifugation in a

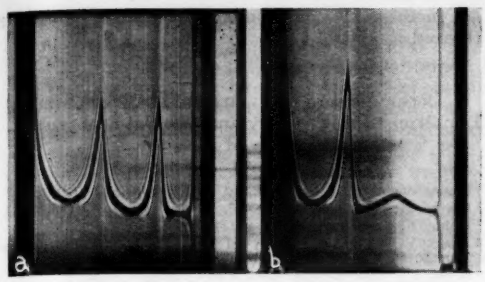

Fig. 1. Schlieren photographs from the 'Spinco' Model *E* ultracentrifuge. Solvent: aqueous carbon dioxide (0·035 *M*, *p*H 5·0). Speed 59,780 rev./min. Sedimentation from right to left. (*a*) Partially dissociated ovalbumin – rabbit antiserum complex. Initial $C = 1·0$ per cent w/v. Exposure after 34 min. at full speed. (*b*) Dissociated horse hæmoglobin – rabbit antiserum complex. Initial $C = 0·85$ per cent w/v. Exposure after 101 min. at full speed

bucket rotor ('Spinco' *SW* 39) at 150,000*g* for 6 hr. Attempts to increase the yield by reworking the deposits were increasingly penalized by the formation of complexes with reduced S_{20} values as more antibody was removed.

The isolated globulin showed no evidence of heterogeneity on sedimentation and electrophoresis, and the absence of antigen was confirmed by observation on antibody dissociated from a complex of ovalbumin labelled with fluorescein. In veronal buffer (*I* 0·1, *p*H 8·6) it had an S_{20} (6·56 S.; $C = 0·3$ per cent) and an electrophoretic mobility ($-1·2 \times 10^{-5}$ cm.^{2} V.$^{-1}$ sec.$^{-1}$) similar to those for normal rabbit γ_2-globulin. Its serological properties appeared to be unimpaired; at least 90 per cent was precipitable by ovalbumin and the characteristics of the precipitin curve were similar to those obtained for the original antiserum.

The work was extended to a specific precipitate of horse hæmoglobin and its rabbit antiserum prepared by a short-term immunization[5]. Once more the precipitate was readily soluble in aqueous carbon dioxide (0·035 *M*), but here a complete dissociation of complex occurred.

Sedimentation analysis of this solution (Fig. 1*b*) showed a major boundary corresponding with free immune globulin and a minor boundary ($S_{20} \simeq 2$) which corresponded with horse hæmoglobin when run under these conditions. The absorption of light (546 mμ) was clearly associated with the minor boundary, but of greater significance was the uniformity of absorption over the major boundary, which indicated that complete dissociation of the complex had been achieved. No hæmoglobin complex appears to exist at *p*H 5·0 in aqueous carbon dioxide, whereas the existence of complexes in solution was demonstrated by sedimentation analysis of specific precipitate dissolved in veronal buffer (*I* 0·001, *p*H 8·3). Here a broad boundary ($S_{20} \simeq 29$) and a trace of globulin were observed but no peak corresponding with hæmoglobin. The light absorption by hæmoglobin was only detectable in the main peak and thus the hæmoglobin was presumably fully complexed.

A cation-exchange cellulose (*CM*)[6] was used instead of the ultracentrifuge to separate the two components of this system, and antibody was isolated in good yield. Aqueous carbon dioxide (0·035 *M*) was used to equilibrate the cellulose column, for the application of the complex and in the development of the chromatogram. Afterwards, antibody was eluted with phosphate-saline buffer (*I* 0·044, *p*H 7·1) in 85 per cent yield while hæmoglobin remained strongly adsorbed. The eluted protein was identical with γ-globulin on paper electrophoresis and in sedimentation analysis, and the absence of hæmoglobin was confirmed by measurements of light at 410 mμ. The product contained at least 55 per cent precipitable antibody despite the susceptibility of the original hæmoglobin system to the formation of soluble complexes. Furthermore, 75 per cent of the added antigen was found in the equivalence zone precipitate in contrast to the low hæmoglobin recoveries (about 18 per cent) in precipitates from the original antiserum.

The work has been extended to the following systems: a polysaccharide[7] from *Shigella dysenteriae f* rabbit antiserum to the homologous *O* somatic antigen, horse serum albumin/rabbit antiserum to horse serum, and diphtheria toxin/horse antitoxin. These specific precipitates dissolved in aqueous carbon dioxide and preliminary examination in the ultracentrifuge indicated that some γ-globulin was released.

The solution and partial dissociation of the ovalbumin complex is not specific to aqueous carbon dioxide. It can be effected to varying extent with many organic and inorganic acids at *p*H 5, and even in the *p*H range 7–8, providing the ionic strength of the solution is low. At present, however, aqueous carbon dioxide appears to be the most advantageous method, and one of the mildest yet reported for dissociating some serological complexes. A full account of this work will be given elsewhere.

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Effect of Benzene on Rat Thyroid

In previous investigations on the anterior pituitary of rats intoxicated with benzene we found histochemical changes suggesting an increased hormone secretion from both the acidophil cells and the beta cells[1]. Since the beta cells are believed to be concerned in the rat with the secretion of thyrotrophic hormone and are therefore also called thyrotrophs[2,3], it seemed of interest to determine whether the hyperfunction of the pituitary beta cells is paralleled by cytological signs of augmented activity in their target gland, the thyroid.

Fourteen male Wistar rats, weighing on an average 260 gm., were given daily subcutaneous injections of 1 ml./kgm. body-weight of a solution containing equal parts (v/v) of benzene and olive oil. Four animals receiving only olive oil served as controls. All rats were killed by gas on the morning following the twenty-second injection. The thyroids were fixed in Helly's fluid and embedded in paraffin.

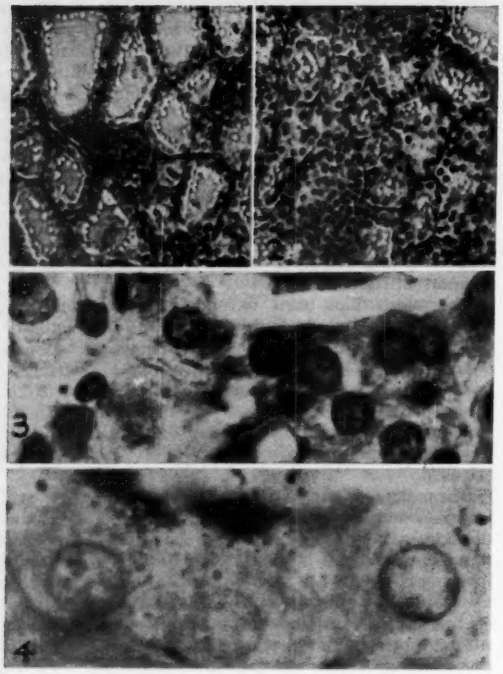

Fig. 1. Normal rat thyroid (hæmatoxylin–eosin). (× 160)

Fig. 2. Treated rat thyroid (hæmatoxylin–eosin). (× 160)

Fig. 3. Showing the cytoplasmic chromidial substance in treated rat thyroid (toluidine blue). (× 1,200)

Fig. 4. Colloid droplets at the apical pole of follicular cells in treated rat thyroid (periodic acid–Schiff). (× 2,000)

Sections were cut at 5μ thickness and stained by one of the following techniques: hæmatoxylin–eosin, methyl green–pyronin, Feulgen reaction, toluidine blue, periodic acid–Schiff reagent.

A series of morphological alterations has been consistently found in all the thyroids of the rats intoxicated with benzene. Most follicles show a much smaller size than those of control rats (Figs. 1 and 2). The follicular cells are high and columnar in shape, their nuclei appear vesicular and weakly stained by Feulgen, and the cytoplasm contains an increased quantity of chromidial substance (ergastoplasm) (Fig. 3), which is apparently related to the capacity of the thyroid cells to synthesize proteins[4,5]. Variable numbers of colloid droplets are clearly demonstrated in the cytoplasm of the follicular cells with the periodic acid–Schiff reaction: they are extremely abundant in some cells and very infrequent in others, while in some cells colloid droplets can be seen only at the apical or at the basal pole (Fig. 4). The colloid accumulated in the lumen of the follicles shows more numerous and larger vacuoles than in control rats, and is generally less intensely stainable (Figs. 1 and 2). Interspersed among the follicular cells lie many parafollicular cells, containing a scanty amount of chromidial substance (Fig. 3) and only few colloid droplets. The interfollicular cells are also abundant (Fig. 2).

Our findings resemble very closely the structural changes which occur in the thyroid when the gland is stimulated[4,5]. Therefore, it seems that under our experimental conditions benzene intoxication causes an increase in the secretory activity of the rat thyroid.

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Separation and Isolation of Fractions of Rabbit Gamma-Globulin containing the Antibody and Antigenic Combining Sites

Some years ago[1] the digestion of rabbit anti-ovalbumin with papain powder was investigated, and it was found that a fragment could be obtained which would no longer form a precipitate with ovalbumin but which would specifically inhibit the precipitation of ovalbumin with undigested antiserum. This fragment, which presumably contained an antibody-combining site, was believed to have a molecular weight of 40–50,000, or about one-quarter that of the original γ-globulin. Earlier, Peterman[2] had shown by ultracentrifuge studies that digestion of human γ-globulin with crude bromelin and papain produced fragments of one-half to one-quarter the size of the original molecule.

In both cases large amounts of crude enzymes were used (50–100 per cent of the weight of the substrate) and consequently investigation of the digestion products was confused by the presence of substantial amounts of non-γ-globulin protein. We therefore decided to repeat the digestion of rabbit γ-globulin with papain, this time using the crystalline enzyme, which can now be prepared[3] easily, and in much lower enzyme-substrate ratios. The conditions of digestion were: 100 mgm. rabbit γ-globulin and 1 mgm. crystalline mercury papain in 3 ml. buffer, *p*H 7·0 (0·1 *M* potassium phosphate, 0·01 *M* cysteine, 0·002 *M* sodium ethylenediamine tetraacetate). This solution was incubated for 16 hr. at 37° C. The digest was dialysed with vigorous stirring and several changes of outer fluid at 2° C. More than 90 per cent of the protein was retained in the dialysis sack and

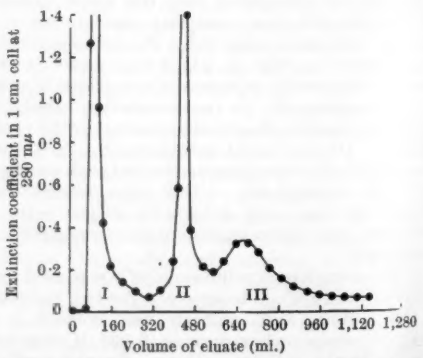

Fig. 1. Fractionation of a papain digest of rabbit γ-globulin by gradient elution on a carboxymethyl cellulose column. Sodium acetate, *p*H 5·5, gradient 0·01–0·9 *M*

when this material was examined in the ultracentrifuge it showed only a single component with $S_{20,w} = 3{\cdot}5$. No effective resolution could be obtained by electrophoresis but on chromatography on carboxymethyl cellulose[4], using gradient elution with an acetate buffer at *p*H 5·5, three components were separated in approximately equal amounts (Fig. 1). Preliminary estimation of the molecular weights of these components by Dr. P. A. Charlwood using the Archibald procedure[5] gave values of 55–60,000. It would thus appear that the rabbit γ-globulin is split into thirds with very little production of small fragments.

If the γ-globulin contained anti-ovalbumin, anti-bovine serum albumin (B.S.A.) or anti-human serum albumin (H.S.A.) fractions I and II specifically inhibited the combination of the same antigen and undigested antiserum, but had no activity in a heterologous system. Thus a digest of anti-human serum albumin γ-globulin will inhibit the human serum albumin – anti-human serum albumin reaction but has no influence on the bovine serum albumin – anti-bovine serum albumin system (Fig. 2). If the γ-globulin contained anti-pneumoccocus polysaccharide type 3 or did not contain any known antibody then these fractions had no activity. The negative result with the polysaccharide antibody may imply a basically different structure to anti-protein antibodies but it may also be a reflexion of the quite different physical character of the antigen and the type of precipitate formed. It is possible that a different technique would show that fractions I and III from an anti-polysaccharide digest also retained the power to combine specifically with the antigen.

Fraction III showed no power to combine with the homologous antigen whatever type of antibody was present in the digest. However, when the antigenic activity of the fraction was tested with a goat anti-rabbit γ-globulin serum it was found that this fraction would precipitate most and probably all the anti-γ-globulin antibody. Fractions I and II neither formed a precipitate with the anti-γ-globulin serum nor did they inhibit precipitation of whole rabbit γ-globulin with the goat anti-serum. Fraction III has a minimum solubility between *p*H 5 and 7·5 and may be readily crystallized and recrystallized by dialysis of an alkaline or acid solution against 0·1 *M* phosphate buffer *p*H 6·5. The crystals (Fig. 3) are diamond-shaped birefringent plates, well formed but thin and easily broken, so that fragments seem always to be present. This ease of crystallization is surprising as whole rabbit γ-globulin has never been crystallized.

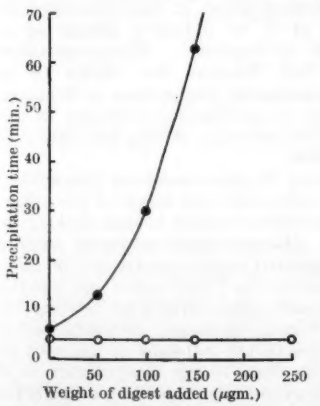

Fig. 2. Inhibition of precipitation of human serum albumin and rabbit anti-human serum albumin by a papain digest of rabbit γ-globulin containing anti-human serum albumin. ●, Precipitation time of human serum albumin – anti-human serum albumin; ○, precipitation time of bovine serum albumin – anti-bovine serum albumin

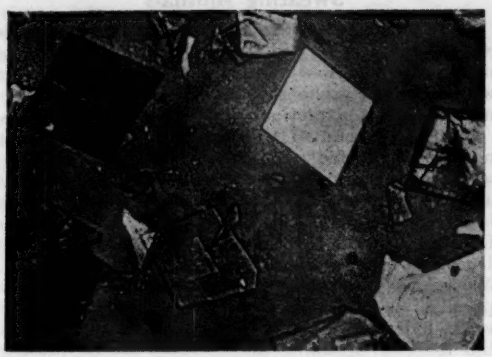

Fig. 3. Crystals of fraction III. Polarized light. (× 50)

These results suggest that the γ-globulin molecule may be made up of three parts; one of these (my crystalline fraction III) is identical in all γ-globulins molecules and is responsible for the common antigenic specificity of γ-globulins observed by Treffers and Heidelberger (see Treffers[6]). The other two parts (fractions I and III) presumably contain the antibody-combining centres, and are also responsible for the physical heterogeneity of γ-globulins; these parts may be expected to vary from molecule to molecule. This suggested structure of an identical section in all molecules of γ-globulins and two variable sections which contain the antibody-combining sites is very similar to the picture drawn by Pauling[7] in 1940. At that time he suggested that the flexibility of the variable sections might be a result of high proline content, but a rough comparison of the amino-acid composition of the three fractions by paper chromatography showed the proline content of each to be of the same order. We have no information on the essential feature of Pauling's theory of antibody formation, namely, that the antibody-combining sites are formed by refolding of a polypeptide chain with an identical amino-acid sequence. The N-terminal pentapeptide is the same in several rabbit antibodies[8,9] but it remains to be shown that this identity of sequence persists through the peptide chain, including the section containing the antibody-combining site.

Fuller details of this work will be published elsewhere.

I wish to thank Dr. P. A. Charlwood for studies of the three fractions with the ultracentrifuge, Dr. J. H. Humphrey for the gift of goat anti-rabbit γ-globulin serum, Mrs. O. Kennard for advice on crystallization techniques and Miss H. Morgan for her technical assistance.

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Heat, Salt and Hormones in Panting and Sweating Animals

In hot climates sweating or panting to provide evaporative cooling are required for the survival of mammals. The loss of extracellular water is linked with cell water exchanges, renal water-saving, adjustments of salt excretion and hormone production.

Within 30 min. of the induction of sweating in normally hydrated man (7 subjects) at 41° C. there was reduction of urine flow to rates below 0·3 ml./min. and reduction in the rates of excretion of sodium below 20 μequiv./min. This was associated with a fall in the sodium/potassium in the urine (Fig. 1*A*). During a period of sweating at the rate of 430–500 ml./hr. conservation of sodium and water ran parallel, as a renal compensation for salt lost in sweat[1]. Ten times the resting amount of aldosterone-like activity appeared in the urine during heating, and this probably accounts for the sodium retention[2] (see also Weiner, J. S., 1st Bioclimatological Cong., Int. Soc. Bioclimatology and Biometeorology, Vienna, September 23–27, 1957; Abstract).

When sheep were studied in a similar hot environment a different pattern emerged. Romney sheep exposed to 41° C. for 4 hr. showed a prompt fall of urine flow on heating. As the respiratory-rate increased for evaporative cooling there was a progressive increase in excretion of sodium and the ratio sodium/potassium rose (Fig. 2). No aldosterone activity was detected in the urine. When sheep were later provided with water to replace that evaporated, the excretion of sodium ceased and there was reversion to a low ratio sodium/potassium in the urine (Fig. 1*B*). Excretion of potassium declined regularly from 200 to 50 μequiv./min. during the period of heating while sodium moved in the opposite direction. Sheep excrete sodium by day more than by night, but the diurnal excretion of sodium is not reversed by heating. The elimination of sodium is presumably a homœostatic compensation for the distillation of water from the respiratory tract. Sheep appear to have no effective sweat glands and are not known to lose electrolytes through the skin during evaporative cooling. Merino sheep were more variable in their responses than Romneys, but followed the same course.

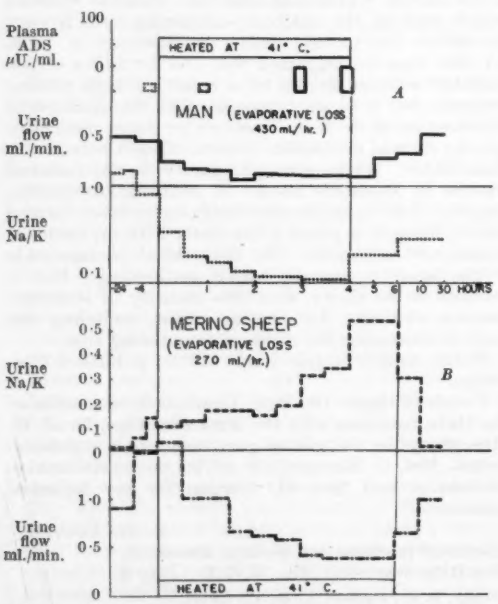

Fig. 1. *A*. Effect of heating at 41° C. on urinary flow and the ratio sodium/potassium in man. The urine flow reached 0·20 ml./min. before antidiuretic substance (ADS) appeared in the plasma. Sodium excretion fell during heating and sweating. *B*, In Merino sheep urinary flow fell on heating but the ratio sodium/potassium and excretion of sodium increased. There was a reversal of the ratio sodium/potassium as cooling took place

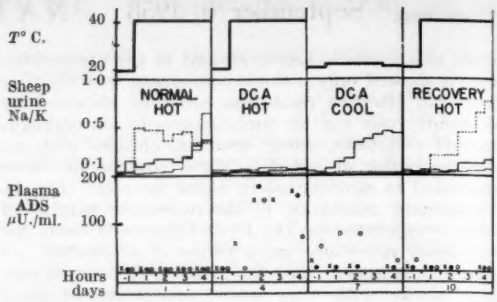

Fig. 2. Romney sheep (3) heated for 4 hr. at 41° C. in winter. No antidiuretic substance (ADS) appeared initially, but large amounts were released into the plasma after injections of deoxycortone (DCA) (0·15 mgm./kgm.). There was a low sodium excretion. Ten days later sodium was excreted during heating, but there was no production of antidiuretic substance

Winter exposure to the standard heat stimulus in both man and sheep led to low rates of flow of concentrated urine (0·1–0·3 ml./min.) without the liberation of measurable quantities of antidiuretic substance into the plasma[3–5]. During summer, antidiuretic substance appeared after 2–4 hr. heating at 41° C. The possibility that salt-active adrenal steroids might regulate the transfer of water and thus the release of antidiuretic substance was tested by exposing three Romney ewes to the standard hot environment in winter, when no antidiuretic substance normally appeared. The ewes were then injected intramuscularly with 15–20 mgm. of deoxycortone and heated for 4 hr. (Fig. 2). Little urinary sodium was excreted but antidiuretic substance appeared in the plasma on heating. As the effects of the hormone wore off the amount of antidiuretic substance appearing at 41° C. was reduced and after 10 days there was virtually no residual activity. In ten Merino sheep it was necessary to heat for 4 hr. at 44° C. to induce a release of antidiuretic substance in summer. When deoxycortone was injected into Merinos the release of antidiuretic substance persisted longer than in Romneys. Winter production of antidiuretic substance in the heat was achieved by injection of this salt-active hormone in both breeds.

In seeking the mechanism of this action of deoxycortone, estimates were made of the plasma volume and extracellular volume of man and sheep subjected to heat[6]. During winter when no antidiuretic substance appeared in the plasma of four men there was no decrease in the *T*1824 space and less than 1 l. loss of thiocyanate space after 4 hr. heating with the loss of 2·5–3·5 kgm. of sweat. Sheep heated for 4–6 hr., with the loss of 1·0–2·0 kgm. of water by evaporation showed less than 10 per cent change in the plasma volume in winter when antidiuretic substance did not appear. In summer, or when deoxycortone was given, there was a reduction in plasma and interstitial

volume during heating and antidiuretic substance was detected in the blood. In the monkey, Sweet *et al.*[7] report an increase of the inulin space on treatment with deoxycortone and in sheep during summer we have found a 25 per cent expansion of the thiocyanate space relative to winter. This would not prevent plasma concentration from occurring if cell permeability to water were affected by the deoxycortone.

It appears that there is a difference between the summer and winter availability of tissue water in both man and sheep. In winter, extracellular water may be mobilized from other sources but not so easily as in summer. Simulation of summer responses to water loss was achieved by injections of deoxycortone. On the other hand, sodium retention in man[8] and excretion of sodium in sheep during heating at any time of the year means that salt-active hormones cannot be effective during the period of heating in both animals. Yet sheep heated in winter mobilize fluid to replenish the plasma space, thus preventing the relative plasma concentration associated with the release of antidiuretic substances, while sodium continues to be excreted. The sodium-resorbing hormone appears, then, not to be the final answer to this problem.

Sodium excretion during heating is thus linked to the method of evaporative cooling. In man producing salt sweat, sodium is retained by the kidney, whereas in sheep, which pant, there is excretion of sodium. There is a seasonal change in the amount of replacement of extracellular water from other sources during heating. When plasma is concentrated, antidiuretic substance appears, and plasma concentration on heating can be induced by deoxycortone. Flow of urine may be reduced to 0·2 ml./min. without antidiuretic substance being detectable in the plasma.

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Correlation between the Fat and the Protein Content of Human Milk

I have undertaken a study of the chemical composition of mature human milk (excluding colostrum) in the Kivu region of the Belgian Congo. The analyses were done on 250 samples of all the milk secreted during 24 hr. by mothers belonging to the Bashi tribe. This ethnic group lives along the eastern shore of Lake Kivu (about long. 29° E. of Greenwich) in the Belgian Congo, between latitudes 2° 10′ S. and 2° 40′ S. at an altitude varying from 1,460 to 2,000 metres above sea-level. Their staple foods are beans, sweet potatoes and beer made from bananas. Demaeyer took the samples in his quantitative study of milk secretion.

I found the mean caloric value, the protein-, fat- and ash-content of the milk from African mothers in the Kivu to be lower, and its lactose-content to be higher (although no statistical analysis of the difference between the means was possible), than the corresponding averages calculated by Macy *et al.*[1] from most literature reports on the composition of human milk and dealing mainly with European and American women.

The observation that both the fat and the protein in human milk are lower here than in other regions with higher dietary standards led us to examine the relation between the fat- and the protein-content of the milk from the mothers here.

A very strong positive correlation was found between the 'protein' (total nitrogen × 5·12)-content and the fat-content of the milk ($n = 224$, $r = 0{\cdot}4426$, $P < 0{\cdot}001$).

This means that for this region a milk sample with a low fat-content tends to have also a low 'protein'-content, that the fat-content of the milk and the 'protein'-content of the milk rise together, and that a milk sample with a high fat-content tends to have at the same time a high 'protein'-content. As far as we know, this correlation has not been observed before in human milk, and we think that it would be important if some of the workers in this field would examine their results in this light, to see whether the same correlation is observed elsewhere, with mothers having a completely different diet. It cannot be calculated from the literature on the subject because only mean values for the various components are published and not the analyses of individual samples. This observation may help towards a better understanding of the relation between the diet of the mothers and the nutritive value of their milk, and perhaps even of the mechanism of the milk secretion in the mammary gland. Breirem[2], in his review of the literature on the relation between the nutritional value of cow's milk and the nutrition of the animal, reports that Mellgaard and Lund, Steensberg, Kay, Breirem and Saarinen have observed that deficiency of energy and/or protein in the diet of the cow will reduce both the fat- and protein-content of the milk. Janz *et al.*[3] report a similar observation made by Escudero and Pierangeli for human milk ; these authors reported that mothers with a diet rich in proteins secreted milk with a high protein- and fat-content.

These reports give indirect circumstantial evidence for the existence of this positive correlation between the fat- and protein-content of milk observed so clearly in my work.

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Effect of Various Natural Steroids on the Phagocytic Activity of the Reticulo-Endothelial System

It was initially observed by Nicol[1] that, following the administration of œstrogenic hormones, there was stimulation of the activity of phagocytic cells in the endometrium. This work was extended later to include both natural and synthetic œstrogens[2–4], and these findings indicated that in both male and female animals œstrogens possess a strong stimulating influence on the reticulo-endothelial system, which results in the appearance of large numbers of macrophages in the spleen, liver and lymph nodes, and the mobilization of these macrophages into the circulation. In the above researches, the trypan blue method was used to assess the activity of the reticulo-endothelial phagocytes. These results have since been confirmed by the carbon method, and it has been found that the strongest stimulants of the reticulo-endothelial system possess high œstrogenicity[5,6].

The following investigation was planned to ascertain the effect of various œstrogenic and non-œstrogenic natural steroids on the phagocytic activity of the reticulo-endothelial system.

The present experiments were carried out on 105 male white mice (*T.O.* Swiss strain) of 20–30 gm. body-weight. Five of the animals were used for assessing the effect of each compound on the phagocytic activity of the reticulo-endothelial system, each animal receiving one subcutaneous injection of 0·5 mgm. of each steroid in 0·05 ml. of arachis oil daily for six days. The phagocytic activity was then measured on the eighth day by the rate of disappearance of a known amount of carbon from the circulating blood[7], the procedure being as follows. Each animal was given one intravenous injection of carbon of particle size 250 A., the dose of the carbon being calculated on the basis of 16 mgm. per 100 gm. body-weight. Blood was then taken at short intervals during the first 60 min. following the injection, and the concentration of the carbon in each blood sample was measured by means of an absorptiometer. The logarithmic values of the absorptiometer readings lie along a straight line when plotted against time, and the slope of this line has been taken as the measure of total body phagocytic activity or the phagocytic index, which is denoted in the present communication by the symbol *K*.

Twenty-five of the animals were used as controls. Each received one subcutaneous injection of 0·05 ml. arachis oil daily for six days and then showed an average phagocytic index or *K*-value of 13 ± 2·4, the carbon completely disappearing from the blood in about 100 min.

Table 1 shows the *K*-values of the various natural steroids together with their relative standard deviations. These results show that the phagocytosis of particulate carbon may be modified by natural steroids. The œstrogenic steroids caused rapid removal of the carbon from the blood-stream due to stimulation of reticulo-endothelial activity. Progesterone caused very slight stimulation of the reticulo-endothelial phagocytes. Cortisone caused marked depression of phagocytic activity, thus providing further confirmation of our previous reports of experiments involving the method of *intravitam* staining with trypan blue[8]. Testosterone and cholesterol and the other steroids shown in Table 1 had little or no effect.

Table 1. Effect of Various Natural Steroids on the Phagocytic Activity of the Reticulo-Endothelial System

Steroid used	Phagocytic index (*K*-value)
Œstradiol benzoate	81 ± 15
Equilin	77 ± 24
α-Ethinyl œstradiol	75 ± 8·9
Œstrone	65 ± 11
Equilenin	51 ± 8·2
Œstriol	47 ± 9·8
Progesterone	17 ± 2·5
Ethisterone	14 ± 1·7
Testosterone	13 ± 1·1
Cholesterol	11 ± 2·5
Lanosterol	11 ± 1·7
Ergosterol	10 ± 1·9
Androstalone	10 ± 1·3
Sitosterol	10 ± 1·1
Deoxycortone acetate	10 ± 2·2
Cortisone	7 ± 0·9
Control values for 25 animals	13 ± 2·4

It is thus evident that the phagocytic activity of the reticulo-endothelial system is modified by hormones, and that the steroids possessing the power of inducing œstrus also induce stimulation of macrophage activity. Œstradiol benzoate has been found to be the most powerful of the natural œstrogens and gives a phagocytic index or *K*-value of 81 ± 15, the carbon disappearing completely from the blood in about 15 min. compared with 100 min. in the controls; cortisone acetate was again the major depressant, the circulating blood not being clear of carbon for about 2 hr. after the injection.

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Action of Beta Radiation on the *Proteus* P18 Bacillus

We have grown the *Proteus* P18 bacillus at 37° C. in an ordinary medium containing 20 per cent horse serum and varying amounts of radioactive phosphorus. With a dose of 4 μc. of phosphorus-32 per 100 ml. of medium the multiplication and microscopic appearance of the bacilli are scarcely altered. When the concentration of phosphorus-32 is raised to 8 μc. per 100 ml., growth is slowed down and a series of important morphological transformations can be observed.

Between 4 and 6 hr. after inoculation, besides the normal *Proteus* bacilli, filaments 20–80μ in length, that show rapid creeping movements, appear (Fig. 1*b*). After 6–8 hr., filaments with a globular formation at one end are observed (Fig. 1*c*); these move very fast, the globule foremost. After about 8–10 hr.

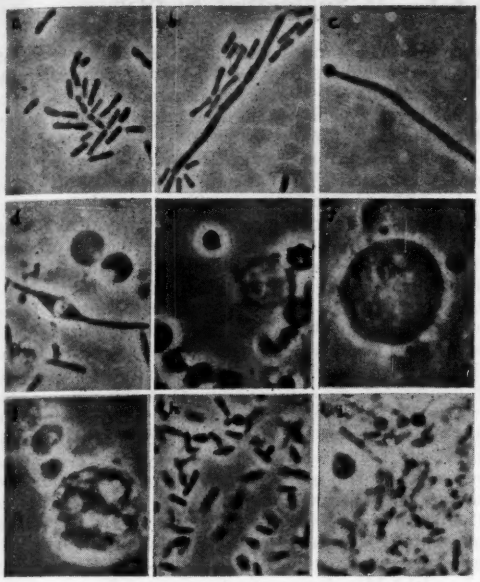

Fig. 1. Morphological changes of *Proteus P*18 under the influence of radiation from phosphorus-32. (*a*) Normal *Proteus*; (1*b*–1*i*) see text

numerous dark globular elements 1–8μ in diameter are seen, as well as filaments with a median bulge (Fig. 1*d*). At the same time the normal bacilli become rare and the long filaments are very numerous. Certain globular formations have a small, very thin and spindly appendage in a spiral (Fig. 1*e*). After 15 hr. bright globular elements 8μ in diameter and small particles at the limit of visibility appear. After about 21 hr. the culture contains scarcely anything but dark or bright globular formations, 30–40μ in diameter, with black granulations or vacuoles (Figs. 1*f* and *g*). The small particles at the limit of visibility are very numerous; but we were unable to photograph them. When the culture is prolonged, the globular formations become fewer and are replaced by a considerable number of small particles. After about 31 hr. there is a regression, and both short and long filaments and many thick rods appear; some of the long filaments have a median bulge. Finally, after 48 hr., one finds normal bacilli again (Fig. 1*h*).

If a 21-hr. culture made in the presence of 8 μc. of phosphorus-32 and rich in globular formations is re-inoculated into a medium containing the same quantity of radioactive phosphorus, one obtains almost exclusively globular formations. A second transplantation under the same conditions after 24 hr. also shows many whole or destroyed globular formations, but mostly rods and short filaments. At the third transplantation the globular formations disappear almost completely and the culture consists of long filaments, rods and normal bacilli. Still more transplantation in the presence of phosphorus-32 produces a more and more normal culture, containing only a few filaments or globular formations.

When *Proteus P*18 develops in a medium containing 16 μc. of phosphorus-32 per 100 ml. of medium, similar changes are observed but they occur slightly faster (Fig. 1*i*).

At a dose of 28 or 40 μc., beside the normal bacilli, one finds a few short filaments and globular formations after 6–9 hr., but neither long filaments nor large globular formations are observable. Apparently the morphological changes are very short-lived and incomplete and the culture quickly reverts to normal. Of course, the larger doses slow down the microbial growth very noticeably; this ceases after about 12 hr.

The transformations which we have described suggest in many respects the *L* forms produced in the presence of penicillin[1,2] by X-rays[3].

The interesting aspect of our experiments is the shortening of the period of unusual morphology when high doses of phosphorus-32 are used, and the reversion to normal forms in spite of the continuous action of radiation. An adaptation to the beta-radiation of phosphorus-32 therefore seems likely.

The metabolism of the bacterial forms produced by phosphorus-32 and its action on other bacilli are under investigation.

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Autonomic Functions of the Hippocampus

At present no definite function can be attributed to the hippocampus. In the cat and the dog, electrical stimulation of the fimbria, the main efferent tract from the hippocampus, has been reported to give rise to no detectable autonomic changes[1], although it is well known that stimulation of other components of the limbic system can elicit autonomic changes.

In rabbits anæsthetized with urethane, pentobarbital sodium or ether, it can be shown that stimulation within the ventral part of the fimbria raises the blood pressure and increases the rate and amplitude of the respiration as illustrated in Fig. 1. Stimulation within the more dorsal part of the fimbria depresses the blood pressure, but has similar effects on the respiration as shown in Fig. 2. The effects obtained from these two regions of the fimbria are qualitatively reproducible in many animals, but vary quantitatively from animal to animal.

Degenerating fibres are being traced by a modified Nauta–Gygax method[2] from lesions made in the fimbria in positions which gave a definite pressor or depressor response when stimulated. In one rat studied by Nauta[3], destruction of the caudal half of the hippocampus led to degeneration of fibres coursing through the middle and ventral parts of the fimbria, some of which continued as the medial cortico-hypothalamic tract to the periventricular zone of the anterior hypothalamus, a region which raises the blood pressure when stimulated. The fimbria is also known to contain fibres terminating in the septum and in the medial mammillary nucleus, both of which give depressor responses when stimulated (Cragg and Hamlyn, unpublished work). It is

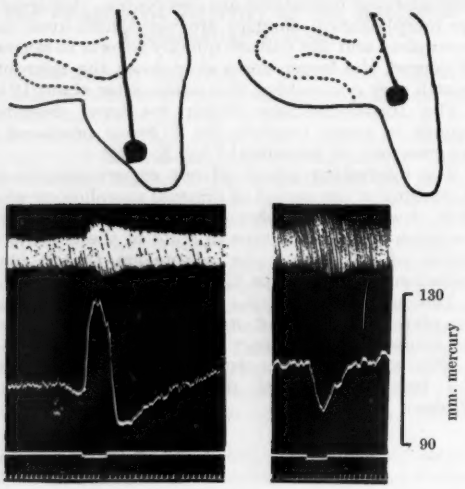

Fig. 1 Fig. 2

Figs. 1 and 2. The changes of respiration and blood pressure produced by stimulation of the fimbria at the positions indicated. Rabbits anæsthetized with urethane and ether. Time marker: 5 sec.

therefore possible that one of the functions of the hippocampus is concerned with the regulation of some of the activities of the autonomic nervous system.

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Semipermeability of the Nuclear Membrane

Previous studies on the osmotic properties of isolated nuclei indicated that ions or molecules of low molecular weight readily penetrate the nuclear membrane. Of the components tested, only protein and certain other substances of high molecular weight were found to exert an osmotic effect[1,2]. The process of isolation may have marked effects on the properties of the nucleus and its membrane. In the present studies, osmotic properties of the nucleus were studied by micro-injection of certain solutions into the cytoplasm. Relatively small, unpigmented oocytes of *Rana pipiens* were used. Photographic records were obtained and all measurements were made from these. No harmful effects were observed within the cells following injection.

In some of the experiments a high molecular weight synthetic polymer, polyvinylpyrrolidone, was used (kindly supplied by Antara Chemicals, General Aniline and Film Corporation, Philadelphia). This substance, which has been used as a blood substitute, has the advantage of being non-toxic, relatively non-reactive, and of known molecular weight. The polymer was dissolved in a solution of potassium and sodium chlorides. For experiments in which potassium and sodium chlorides alone were used, the solution consisted of nine parts 0·126 *M* potassium chloride and one part 0·123 *M* sodium chloride. For the other solutions the concentrations of potassium and sodium chlorides were adjusted slightly so that all solutions had exactly the same tonicity with respect to the cell membrane (isosmotic with amphibian Ringer). The *p*H, in each case, was adjusted to approximately 6·5.

For each solution a series of about ten experiments was performed. The results indicate clearly that polyvinylpyrrolidone has an effect on nuclear volume. The injection of 0·5 per cent of the polymer causes a significant increase in volume of the nucleus, whereas 2·0 per cent or 4 per cent causes a significant decrease. 1 per cent and 1·5 per cent polyvinylpyrrolidone have very little effect on nuclear volume. If we assume that the observed volume changes are osmotic in nature, then nuclear volume is apparently dependent on the colloid osmotic pressure of the cytoplasm. Furthermore, we can conclude that the isotonic concentration of polyvinylpyrrolidone lies in the range 1–1·5 per cent. The molecular weight of this preparation of polyvinylpyrrolidone (type *NP*–K30) is reported by the manufacturer to be 40,000, and the molar concentration for the isotonic solution is therefore equal to $2{\cdot}5 - 3{\cdot}75 \times 10^{-4}$ *M*. In further experiments crystalline bovine serum albumin (molecular weight, 65,360)[3] was used. A 1·96 per cent (3×10^{-4} *M*) solution of albumin was found to cause no significant change in volume of the nucleus. An albumin solution of twice that concentration, however, caused a significant decrease in volume. Injection of a solution of potassium chloride and sodium chloride alone, which is isotonic with respect to the cell membrane (isosmotic with amphibian Ringer), as well as a solution 20 per cent more concentrated, invariably caused an increase in nuclear volume. It might be re-emphasized that the potassium chloride–sodium chloride solutions of polyvinylpyrrolidone and albumin which are hypertonic for the nucleus are isotonic with respect to the cell membrane.

These results would indicate that the nuclear membrane of the intact oocyte does not allow the free penetration of molecules of 40,000 molecular weight and larger. It is possible that some penetration does occur, but to such a small extent that it is not detectable under the experimental conditions employed. There has been no systematic attempt to determine a threshold range of molecular weight, below which the molecules would be osmotically ineffectual. Previous work with this species indicated that sucrose, with a molecular weight of 342, readily penetrates the isolated germinal vesicle nucleus, whereas egg albumin, with a molecular weight of approximately 34,500, does not[2]. On the other hand, it has been shown that blood proteins rapidly get into the nuclei of mammalian somatic cells, where they may even accumulate[4]. It would be important to show if differences exist between the nuclei of germ cells and somatic cells (or even between the same cell types of two different species) that might account for this apparent discrepancy. (See Anderson[5] for a discussion of this.) It is conceivable that active processes are involved in the passage of certain high molecular weight substances (proteins) between nucleus and cytoplasm. The presence of such an active mechanism might allow the penetration of certain large molecules through the nuclear membrane without necessarily destroying the properties of the nucleus as an osmometer. This possibility might reconcile the observations on osmotic properties of nuclei, and the apparent permeability of certain somatic cell nuclei to proteins, as has been suggested by Stern and Mirsky[6].

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Kinetics of Muscular Contraction in Heavy Water

VARIOUS lines of evidence, for example, involvement of sulphydryl groups and adenosine triphosphatase, suggest that the rate-limiting process for muscular contraction may be proton transfer. In this event, one would expect the rate of either isometric or isotonic contraction to be slowed up in the presence of heavy water (D_2O). According to a recent analysis[1], which of the two most affected will depend on whether activation or production of tension is involved.

Fig. 1 shows the development of isometric tension in normal and heavy water – Ringer solution after 20 min. equilibration. The reduction in rate of just over three times is completely reversible. Maximum tension and relaxation phase were not altered; action potentials in another muscle (frog sartorius) were found to be normal. These results, with the exception of 30 per cent less tension, are reproduced by normal Ringer solution with osmotic pressure increased by addition of sucrose to 1·6 times (that is, 0·416 osmolar)

Fig. 2 shows force–velocity curves for the three cases. The isotonic rate is reduced some twelve times in heavy water – Ringer solution, while for sucrose Ringer the factor is again only three.

From this we should conclude that the reaction affected is tension production, and from the magnitude of the effect more than one (possibly three) protons are involved.

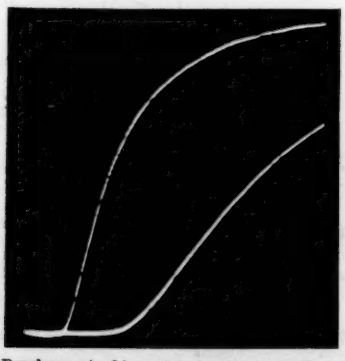

Fig. 1. Development of isometric tension in turtle (*Chrysemys picta*) retractor penis muscle at 0° C. Left to right, in normal and heavy water – Ringer solution

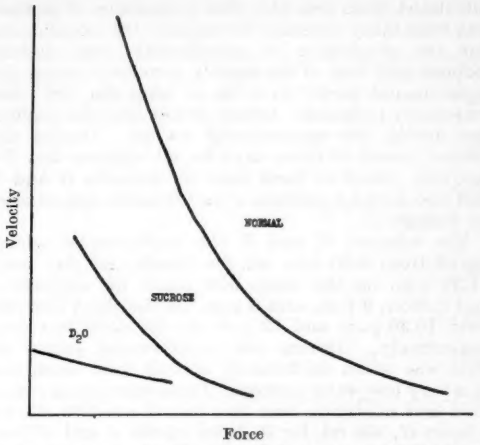

Fig. 2. Force–velocity curves of the same muscle, obtained with a continuously recording device

I am indebted to Dr. N. Geschwind for observation of action potentials by micro-electrode technique, and to the Muscular Dystrophy Associations of America for support.

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Influence of Water Deprivation on Taste Sensitivity in Man

IN a previous communication[1], I presented results showing that forced loss of salt from the body is accompanied by increases in taste sensitivity to 'salt' in human subjects. Sensitivity to 'sweet', 'sour' and 'bitter' remained unchanged. In a study of the preference behaviour of thirsty (deprived of water for 17½–19 hr.) and non-thirsty rats, Young and Falk[2] report that thirsty rats reject hypertonic solutions and tend to prefer water to hypotonic salt solutions. Thus it appears that the preference behaviour of rats deprived of water is practically the opposite of that shown by rats deficient in salt. In view of the finding that salt deficiency is accompanied by an increase in sensitivity to salt, Young and Falk's results suggest that water deprivation may be accompanied by a decrease in sensitivity to salt.

To check this, four human subjects (*H*, *R*, *J* and *C*) were rendered water-deficient and their sensitivity to 'salt' (sodium chloride) and 'sour' (sulphuric acid) was studied. Details of the procedure used to establish absolute thresholds were reported previously[1]. After rinsing the mouth, subjects were careful to spit out as much as possible of the fluid before swallowing. Nevertheless, very small amounts of fluid were undoubtedly swallowed during testing.

Throughout the control, experimental and recovery periods of the experiment (nine days for subjects *H* and *R* and eight days for subjects *J* and *C*) the overall caloric value of the food intake of all subjects was controlled. It averaged about 2,100 cal./day for *H*, about 2,650 cal./day for *R*, about 2,840 cal./day

for *J* and about 2,450 cal./day for *C* (all food was weighed; nutritional values and water content were calculated from ref. 3). The proportion of protein was kept fairly constant throughout the experiment, but the proportion of carbohydrate was slightly reduced and that of fat slightly increased during the experimental period in order to keep the 'dry' diet reasonably palatable. Intake of salt was also slightly less during the experimental period. During the control period of three days for all subjects and the recovery period of three days for subjects *H* and *R* and two days for subjects *J* and *C* water was allowed *ad libitum*.

For subjects *H* and *R* the experimental period lasted from 8.30 a.m. on the fourth test day until 11.30 a.m. on the sixth test day; for subjects *J* and *C* from 9 p.m. and 6 p.m. on the third test day until 10.30 p.m. and 12 p.m. on the sixth test day, respectively. During the experimental period no fluid was taken deliberately and all food eaten was of a very low water content. Total water intake from food and oxidation over this period was 659 ml. for subject *H*, 834 ml. for *R*, 1,000 ml. for *J* and 837 ml. for *C*. Subjects *H* and *R* carried out a 2-hr. sweating session (in a hot room at 95° F. wet bulb, 105° F. dry bulb) during the morning of the first day of water deprivation. Losses in body-weight, recorded over the experimental period, were 2·500 kgm., 3·150 kgm., 2·250 kgm. and 2·900 kgm. for subjects *H*, *R*, *J*, and *C* respectively. Increases in the concentrations of sodium in the serum and chloride in the plasma and in hæmatocrit percentages also occurred in all subjects (see Table 1).

The means of thresholds for 'salt' of subjects *H* and *R*, determined daily at 9.00 a.m. and 4.45 p.m., and of subjects *J* and *C*, determined daily at 9.00 a.m. and 11.30 a.m., are presented in Fig. 1. The means of thresholds for 'sour' of subjects *H* and *R*, determined daily at 2.15 p.m., and of subjects *J* and *C*, determined daily at 2.30 p.m., are also shown in Fig. 1.

Table 1. Concentrations of Sodium in Serum, Chloride in Plasma and Hæmatocrit Percentages for Subjects *H*, *R*, *J* and *C**

Day of experiment	Serum–sodium (m.equiv./l.)		Plasma chloride (m.equiv./l.)		Hæmatocrit (per cent)	
	R	*H*	*R*	*H*	*R*	*H*
3	147	147	109	112		
6	160	155	118	115	48·7	44·9
7	148	146	107	111	48·4	44·8
9	148	144	108	110	46·5	40·9
	J	*C*	*J*	*C*	*J*	*C*
3	144	137	109	104	50·1	45·1
6	147	141	111	113	51·9	46·7
8	144	141	110	108	49·2	46·1

* Thanks are due to Prof. R. A. McCance, Department of Experimental Medicine, University of Cambridge, for arranging the taking and analysis of these samples.

From these results, it can be seen that loss of body-water was accompanied by a decrease in sensitivity to 'salt', whereas sensitivity to 'sour' remained unchanged.

The results also show that sensitivity to 'salt' was less acute at 9 a.m. than at the later session on the same day. I have found evidence of similar systematic fluctuations in sensitivity to 'salt', 'sweet', 'sour' and 'bitter' solutions during the course of a day of normal food intake (to be reported elsewhere). The variations are statistically significant. The degree of variation appears to depend largely on the caloric value and composition of the food intake. Systematic variation during the day in taste sensitivity has also been reported by others, namely, Hammer ('sour'), Goetzl *et al.* ('sweet') and Irvin and Goetzl ('salt')[4]. In the present results, the degree of variation from 9 a.m. to times later in the day has not remained constant during the period of decreased sensitivity to salt. This is probably due to the fact that the tendency for sensitivity to increase from the 9 a.m. session to the later session was partly counteracted by the tendency for it to decrease due to the increasing loss of body-water over this period. It appears, then, that the factors responsible for the changes in taste

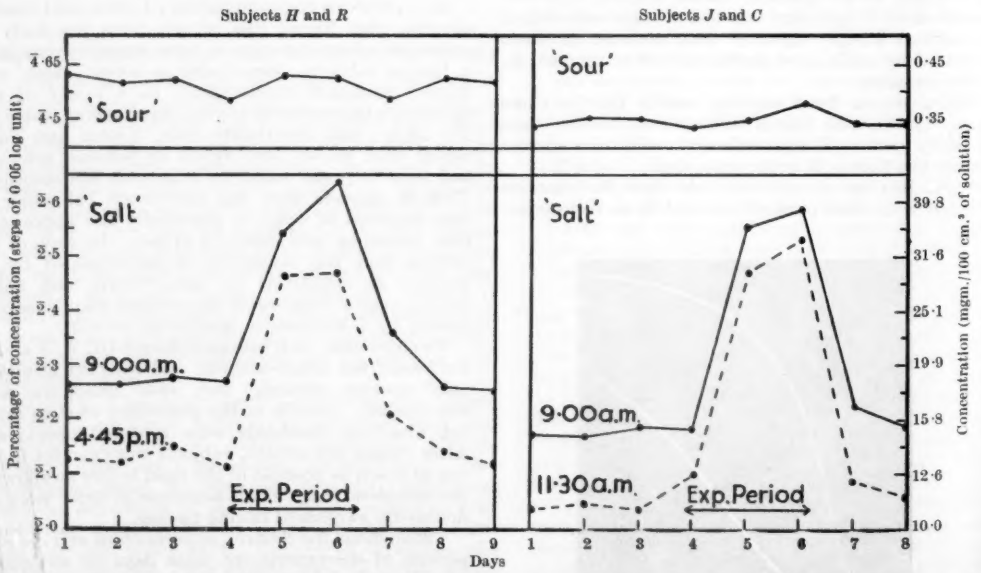

Fig. 1. Mean absolute thresholds for 'salt' and 'sour' for subjects *H* and *R* (left) and for subjects *J* and *C* (right). Each threshold is the mean of eight determinations, four from each of two subjects

sensitivity which occur during the day are independent of the factors causing the observed changes in sensitivity which were specific to 'salt'.

Since losses in body-salt, which result in a fall in the concentration of sodium in the extracellular fluid, were accompanied by increases in sensitivity to salt, and losses in body-water, which result in a rise in the concentration of sodium in the extracellular fluid, were accompanied by a decrease in sensitivity to salt, it appears likely that these changes in sensitivity are related to the concentration of sodium, and possibly the osmotic pressure, of the extracellular fluid (the osmotic pressure is usually, but not always, dependent on the concentration of sodium in the extracellular fluid). Changes in the volume *per se* of the extracellular fluid would not appear to be related to the sensitivity changes, since under both conditions reductions occurred in the volume of the extracellular fluid.

This investigation is made possible by the grant of a Hackett studentship from the University of Western Australia, for which special thanks are due.

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Nitrite Accumulation in the North-East Tropical Pacific

DURING two oceanographic expeditions in waters off the Mexican and Central American coast, nitrite analyses were made on water samples from between the surface and 800 m. depth in order to obtain information on the decay process of planktonic material.

Because of the intermediate position of nitrite in the nitrogen cycle between ammonia and nitrate, its concentration is determined by the ratio of ammonia oxidation to nitrite oxidation, in case of nitrification. The bacteria responsible for the oxidation of ammonia do not occur freely suspended in the open ocean but are attached to plankton organisms. Dying plankton often accumulates in the thermocline, and in culture experiments large quantities of nitrite appear soon after the cultures die[1]. Many investigators have found a nitrite peak associated with the thermocline layer. In highly productive, tropical areas[2] conditions are especially favourable for a strong decay of planktonic material. As a result of this decay all 20 stations occupied 50–650 miles off the American coast from the Gulf of Panamá (4° N.) to the southern tip of Baja, California (23° N.), show this nitrite accumulation in the thermocline layer (Fig. 1).

Denitrifying bacteria also exist in the sea, some of which are able to reduce nitrate to nitrite under anaerobic or nearly anaerobic conditions in the presence of abundant organic matter. In most areas of tropical oceans there is, even in the oxygen minimum layer, some oxygen present. Wattenberg[3] reported 0·37 cm.³/l. as the lowest oxygen value measured on the *Meteor* Expedition in the South Atlantic. Due to the weaker subsurface circulation in the Pacific and a relatively high production in its eastern part, a large body of water is found within which the oxygen content is almost zero[4]. This water of low oxygen content extends off the American coasts between 8° N. and 21° N. in an intermediate layer between 150 and 1,000 metres. Fig. 1 reveals that only the area with very low oxygen content is also a region with a second, deep nitrite maximum, with values up to 2·5 μgm. at./l. Both features, low oxygen and high nitrite concentrations, are found between 150 and 500 metres.

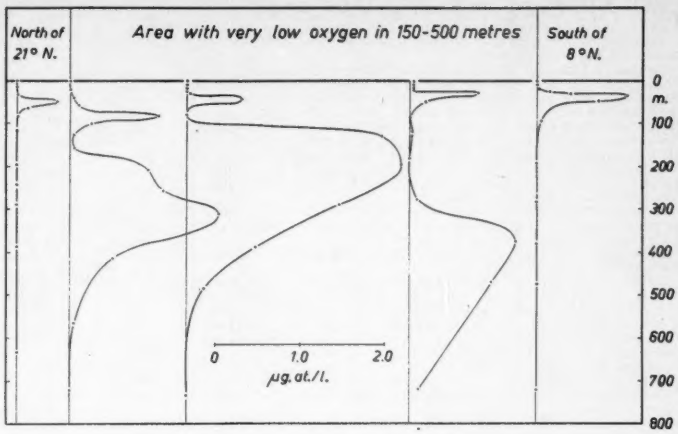

Fig. 1

From an oxygen versus nitrite plot the two nitrite maxima are clearly distinguishable. Nitrite from the upper maximum is found within a wide oxygen range from about 1 to 5 cm.³/l., due to the very sharp thermocline with which this maximum is associated. The nitrite values of the deep maximum occur only in water with less than 0·10 cm.³/l. oxygen, which suggests that the deep nitrite maximum is produced by reduction of nitrate to nitrite. Similar observations of a deep nitrite maximum were made by Gilson[1] in the eastern Arabian Sea, but at that time could not be explained.

This work was partly supported by funds of the Inter-American Tropical Tuna Commission, Scripps Institution of Oceanography, La Jolla, California. A detailed report of this study will be published elsewhere.

WILHELM BRANDHORST

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der Universität Kiel. June 18.

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